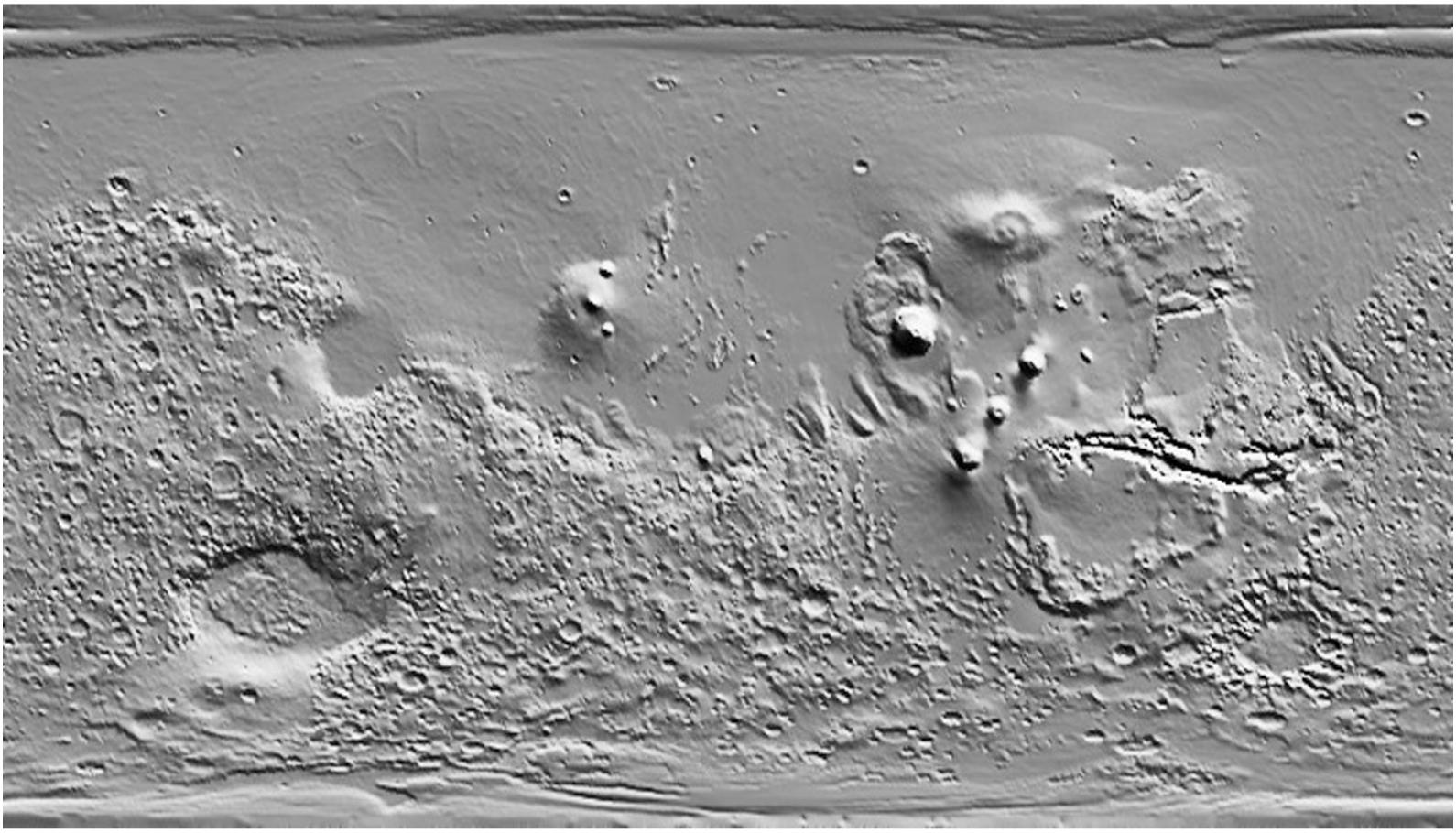
Mars Astrobiology Science and Technology Workshop Abstracts

8-10 September 2004
Carnegie Institution of Washington
Washington, DC



Sponsored by: the NASA Office of Space Sciences, the NASA Astrobiology Institute, the Carnegie Institution of Washington, and the NASA/JPL Mars Program Office.

Meeting Co-Chairs: Bruce Jakosky (Univ. of Colorado), Greg Bearman (JPL), and Andrew Steele (Carnegie).

Organizing Committee: Greg Schmidt (NASA/Ames), Michael Meyer (NASA HQ), David Lavery (NASA HQ), Karen McBride (NASA HQ), Tim Krabach (NASA/JPL), Shirley Berthold (NASA/Ames), Sharon Bassin (Carnegie)

Workshop on Mars Astrobiology Science and Technology

Carnegie Institution of Washington
(1530 P St., NW, Washington, DC, two blocks from DuPont Circle, main auditorium; enter building through the P St. entrance)

8-10 September 2004

Sponsored by NASA Office of Space Sciences, NASA Astrobiology Institute, and Mars Exploration Program Analysis Group (MEPAG).

(Final agenda, 20 August 2004)

Note on posters: All posters should be up for the entire meeting, and each will be highlighted during the designated session.

Tuesday, 7 September

7:00-11:00 p.m. Student ice breaker (but all are welcome), location "Buffalo Billiards", 130 19th St., NW, Washington DC.

Wednesday, 8 September

8:00	Registration, continental breakfast	All
8:30	Welcome and introduction to meeting	B. Jakosky G. Bearman A. Steele
8:45	Mars program status	D. McCuistion
9:05	Mars program science status	J. Garvin
9:25	NASA HQ perspective on astrobiotechnology, ASTEP, and ASTID programs	M. Meyer
9:50	PIDDP, MIDP, and SRLIDAP programs	M. Lindstrom
10:05	Break	All
10:20	MEPAG science goals and objectives (emphasizing life)	A. Steele

10:50	Lessons learned from MER about *in situ* quantitative analysis	D. Des Marais
11:20	Planetary protection issues and status	J. Rummel
11:45	ITAR rules and regulations	J. Hall
12:15	Lunch	
1:30	Session 1: Sample handling and planetary protection for in situ analysis and sample return (Discussion leader: Luther Beegle)	
1:30	Introduction to discussion questions	
2:00-3:00	Poster viewing for this session (includes break) - Posters on sample access, handling and planetary protection for in situ analysis and sample return - Posters on technology demos, ASTEP projects, and flight-ready instrumentation	
2:45	Focused discussion on: - Sample concentration methods - Cross-contamination issues - Organic contamination detection and mitigation - Sterilization techniques and instrument survival - Completely clean blanks - Sample extraction - Handling icy and ice samples - Down-hole sample transport and measurements	
4:30	Summary issues and recommendations	
5:00-7:00	Reception	

Thursday, 9 September

8:00	Continental breakfast	
8:30	Session 2: Targeting *in situ* analysis, sample selection, and context measurements (Discussion leader: Pan Conrad)	
8:30	Introduction to discussion questions	
9:00	Poster viewing for this session (includes break)	
10:30	Focused discussion on: - On-surface remote-sensing measurements for target selection - "Contact" imaging at all spatial scales - Microscopic imaging spectroscopy - Imaging elemental and chemical analyses	
11:45	Summary issues and recommendations	
12:00	Lunch	
1:15	Session 3: Microfluidics and biomolecular techniques for detecting biosignatures (Discussion leaders: John Dzenitits and Josh Molho)	
1:15	Introduction to discussion questions	
1:45	Poster viewing for this session (includes break)	
3:15	Focused discussion on: - Microfluidic devices (and how to clean them) - Sensors from bioterrorism developments - Ultrasensitive instrumentation--optical assays, mass spectrometry and nano-mems devices - MicroTAS (total analytic systems)	
4:30	Summary issues and recommendations	
5:00	Student-led discussion on graduate education and educational outreach	K. Lynch L. Taylor
6:00	Adjourn	

Friday, 10 September

8:00 Continental breakfast

8:30 Session 4: Other organic molecule detection techniques
(Discussion leaders: Tony Ricco and Greg Kovacs)

8:30 Introduction to discussion questions

9:00 Poster viewing for this session (includes break)

10:15 Focused discussion on:

- Biomedical sensors
- MicroTAS (total analytic systems)
- Instrument integration issues
- Downhole instruments
- Instrument suite designs

11:30 Summary issues and recommendations

12:00 Meeting wrap-up (with emphasis on gaps in technology development)

12:30 Adjourn

ABSTRACTS SUBMITTED, BY SESSION

Session 1a. Sample access, handling, and planetary protection for in situ analysis and sample return.

The Ultrasonic/Sonic Driller/Corer (USDC) as a Subsurface Drill, Sampler and Lab-on-a-Drill for the Mars Astrobiology Science and Technology, Y. Bar-Cohen, S. Sherrit, X. Bao, Z. Chang, M. Badescu, B. Kennedy, P. Doran, D. Blake, P. Conrad, G. Bearman, and I. Kanik.

The Inchworm Deep Drilling System For In Situ Investigations of Martian Subsurface Aquiferous Zones, T. Myrick, S. Frader-Thompson, J. Wilson, and S. Gorevan.

A Comprehensive Plan for Drilling in Martian Permafrost in Search of Life, H.D. Smith and C.P. McKay.

Subsurface Sampling and Sensing using Burrowing Moles, C.R. Stoker, L. Richter, and W.H. Smith.

Sample Preparation Systems Development for the Microarray Assay for Solar System Exploration (MASSE) Project, M. Potter, K. Showalter, and M.D. Fries

Session 1b. Technology demos, ASTEP projects, and flight-ready instrumentation..

Tools for Detection of Phototrophic and Chemosynthetic Microbial Life on Mars, G. Ananyev, T.C. Onstott, and G.C. Dismukes.

The Mars Astrobiology Probe: A proposed instrument suite for the 2009 Mars Science Laboratory (MSL), J.L. Bada et al.

Molecular Recognition Sensing Materials within Operational and Mission Environments for Planetary Exploration: Consideration of Stability and Robustness, D. Cullen, O. Henry, S. Piletsky, D. Thompson, N. Bannister, M. Sims, S. Nissen and A. Porter.

MEMS-based Force-Detected Nuclear Magnetic Resonance Spectrometer system for Astrobiological Investigations, T. George, K. Son, C. Lee, P. R. Nilsson, R.A. Elgammal, and D.P. Weitekamp.

Mars Analog Rio Tinto Experiment(MARTE): An experimental demonstration of key technologies for searching for life on Mars, C. Stoker.

Field Tests of the Mars Oxidant Instrument (MOI), A.P. Zent, R.C. Quinn, F.G. Grunthaner, and P. Ehrenfreund.

In-Field Testing of Life Detection Instruments and Protocols in a Mars Analogue Arctic Environment, A. Steele, M. Schweizer, H.E.F. Amundsen, and N. Wainwright

Enzyme-Cascade Analysis of the Rio Tinto Subsurface Environment: A Biosensor Development Experiment, K. Lynch, N. Wainwright, A. Child, K. Williams, D. McKay, R. Amils, E. Gonzalez, and C. Stoker

Session 2. Targeting in situ analysis, sample selection and context measurements.

Using Wind Driven Tumbleweed Rovers to Explore Martian Gully Features, J. Antol and J.L. Heldmann.

Tumbleweed: A Wind-Propelled Survey Vehicle for Astrobiology and Planetary Science, K.R. Kuhlman, A.E. Behar, J.A. Jones, F. Carsey, M. Coleman, G. Bearman, M. Buehler, P.J. Boston, C.P. McKay, L. Rothschild, J. Antol, G.A. Hajos, W.C. Kelliher, I.A. Carlsberg, J.P. Keyes, M. Rudisill, and R.L. Crawford.

Astrobiological Applications of the Mbari Environmental Sample Processor (ESP), S.M. Feldman, C. Scholin, J. Feldman, S. Jensen, B. Roman, C. Preston, V. Orphan, and J. Dzenitis.

Astrobiological Significance of Definitive Mineralogical Analysis of Martian Surface Samples Using the Chemin XRD/XRF Instrument, S.M. Feldman, D.F. Blake, P. Sarrazin, D.L. Bish, S.J. Chipera, D.T. Vaniman, and S. Collins.

The 2007 Phoenix Mars Scout MECA Wet Chemistry Laboratory, S.P. Kounaves, M.H. Hecht, P. Smith, and the Phoenix Team.

Thermal Properties of Saline Environments: Potential targets for detecting signatures of past or present life on Mars, S. Scher.

In-Situ Measurements of Ionic Motion Directly in Planetary Soils, S. Seshadri, M.G. Buehler, and R.C. Anderson.

Poor Preservation Potential of Organics in Meridiani Planum Unit P2 Sedimentary Rocks, D.Y. Sumner.

Atmospheric Electron-Induced X-ray Spectrometer (AEXS) Development, J.Z. Wilcox, E. Urgiles, and T. George.

Session 3. Microfluidics and biomolecular techniques.

Using Protein-DNA Chimeras to Detect and Count Small Numbers of Molecules, I. Burbulis, K. Yamaguchi, R. Carlson, and R. Brent.

Remote Spectral Imaging of Geologic Formations and Zoetic Residues with Active Excitation Diode Arrays, R.A. Lodder and G.V. Levin.

Multi-Layer Microfluidic Devices for Amino Acid Biomarker Analysis: The mars organic analyzer, A.M. Skelley, J.R. Scherer, A. Aubrey, P. Ehrenfreund, J.L. Bada, F.J. Grunthaner, and R.A. Mathies.

SMILE (Specific Molecular Identification of Life Experiment): A family of molecular recognition sensor instruments for robust detection of life in the solar system, M. Sims and D. Cullen.

Feasibility of Enzymatic Assay for ATP as an Indicator of Subterranean Microbial Life on Mars, A.W. Szumlas, F. Andrade, L.M. Pratt, and G.M. Hieftje.

Microfluidic Measurement of Microbial Bioburden by Fluorescent LAL Assay, N. Wainwright, N.R. Symonds, W. Nutter, D. Child, A. Lycans, S.N. Monaco, and L. Monaco.

Searching for Life on Mars and Beyond: Using fluorescence biological analysis (FBA) to examine the subsurface of Europa for signs of life, L.T. Taylor, T. Gary, L. Myles, O. Prieto-Ballesteros, J. Gómez-Elvira, D. Fernández-Remolar, F. Gómez, V. Parro, and R. Amils.

Optical Sensors for Biomolecules Using Nanoporous Sol-Gel Materials, J. Fang, J.C. Zhou, E.H. Lan, B. Dunn, P.L. Gillman, and S.M. Smith.

Activity of the Enzyme Hydrogenase as a proxy for Hydrogen Metabolism in Deeply buried Sediments, and as a Diagnostic Test for Life, B. Soffientino and A.J. Spivack

Magnetoelectronic Microarray Detection of Magnetically Labeled Biomolecules, C. Tamanaha, C. Cole, S. Mulvaney, J. Rife, and L. Whitman.

Modular Assays for Solar System Exploration, A.Steele, J. Maule, J. Toporski, L. Monaco, S. Spearing, R Avci, M Schweitzer and N. Wainwright

Lab on a Chip Application Development for Exploration, L. Monaco, S. Spearing, A. Jenkins, W. Symonds, D. Mayer, E. Gouldie, N. Wainwright, M. Fries, J. Maule, J. Toporski, and A. Steele

Unit Operations and Application of Microfluidic Systems for Remote Analysis, A.J. deMello and B.M. Stone.

Session 4. Other organic-molecule-detection techniques.

Carbon and Silicon Nanowire Chemical Sensors for In Situ Astrobiological Measurements, B. Hunt, M. Bronikowski, A. Fisher, and E. Wong.

Technical Approaches to Laser Mass Spectrometry at Mars, W. Brinckerhoff, T. Cornish, S. Ecelberger, S. Jaskulek, J. Boldt, and K. Strohbehn.

High Resolution Electron-Induced XUV Fluorescence for Life Detection, S. Brotton, J. Ajello, J. Wilcox, and J. Guo.

Solvent Extraction and Chemical Derivatization of Organic Molecules of Exobiological Interest for In Situ Analysis of the Martian Regolith, A. Buch, D.P. Glavin, M. Cabane, and P.R. Mahaffy.

Measurement of Microbial Activity in Soil or Atmospheric Dust by Colorimetric Observation of In Situ Dye Reduction: An approach to detection of extraterrestrial life, R. Crawford, I. Erwin, L. Allenbach, and B. Barnes.

Wet Chemistry Experiment at Mars (WetChem), I. Kanik, L.W. Beegle, S. Kounaves, B. Laughlin, R.G. Cooks, M. Hecht, and P.V. Johnson.

Dielectric Spectroscopy for In Situ Detection of Microbial Life Forms on Mars, D. Warmflash, J.H. Miller, Jr., D.S. McKay, G.E. Fox, and D. Nawarathna.

TOOLS FOR DETECTION OF PHOTOTROPHIC AND CHEMOSYNTHETIC MICROBIAL LIFE ON MARS

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An evaluation of the highest probability approaches most suitable for revealing microbial life on Mars is the main focus of this project. Two signatures of microbial life are proposed as targets for characterization in future Mars rover missions: 1) all natural photosynthetic life on earth exhibits a time-dependent adaptation to illumination over the micro-milli-second interval that serves as a unique signature of phototrophic life, and 2) the inter-conversion of disequilibrium in redox potentials of environmental constituents serves as the principal chemosynthetic energy source in the metabolism of heterotrophic bacteria. A top priority of our work is the development of tools for the unambiguous detection and quantitative estimation of these markers on Mars.

A laser-based fluorescence induction spectrometer suitable for remote sensing (100 meters radius via telescope) of variable fluorescence and luminescence emission intensity and spectrum (near UV through near IR) from organic chromophores including photosynthetic pigments and organophosphates (nucleic acids, ATP, FAD, NAD, NADP and their degradation products) is proposed. This scouting tool shall increase the local prospecting probability by enabling a rover to prescreen candidate sites for detailed sample acquisition and analysis. Sampling of ice, rock and airborne particles will be employed. For airborne particles, natural fractionation occurs in air that separates particles of light organic matter from dense mineral phases and shall be employed for enrichment of light matter suitable for high sensitivity analysis. The detection of temporal changes in fluorescence emission intensity upon changes in light excitation intensity (light adaptation) will be an integral part of this spectrometer. Microbial organisms respond to changes in electrochemical potential by altering the redox state of their constituent energy cofactor molecules. This change has been detected in phototrophs using light to initiate redox change and fluorescence emission to detect the redox state (variable fluorescence). It has a characteristic time/light response that differs from abiotic fluorescence or luminescence and is a unique signature of life. Two prototypes of the above spectrometer have been built and tested. The proposed instrument would be a third generation instrument suitable for Mars missions.

Microbes are masters at exploiting gradients in chemical potential to derive energy for growth. They do this by inter-conversion of molecules that have different chemical potentials owing to their different chemical structures or concentration gradients across interfaces. The atmosphere of Mars is exposed to high levels of UV radiation producing O_2 and radical O gaseous species. At the Martian surface these species create an oxidized layer of unknown extent, but a substantial vertical gradient in this electrochemical potential must exist. Measurement of the magnitude and chemical form (speciation) of this O_2 /redox disequilibrium using ultra-sensitive electrochemical and fluorescence techniques suitable for measurement of O_2 concentrations shall be compared. Supported by NAI.

USING WIND DRIVEN TUMBLEWEED ROVERS TO EXPLORE MARTIAN GULLY FEATURES

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Gully features on the slopes of numerous Martian crater walls, valleys, pits, and graben are of particular interest because of their apparent young age and the potential association with liquid water. The overarching question is how did these gullies form? Specifically, what is the agent of erosion and what is the source of the erosional agent? Several mechanisms for gully formation have been proposed, including: liquid water aquifers (shallow and deep), melting ground ice, snow melt, CO_2 aquifers, and dry debris flow. Observational tests conducted using remote sensing by the Mars Global Surveyor - Mars Orbiter Camera (MGS MOC), the Mars Orbiter Laser Altimeter (MOLA) and the Thermal Emission Spectrometer (TES) indicate that the most likely *erosional agent* is liquid water; however, debate concerns the source of this water. The observations favour a liquid water aquifer as the primary candidate and because the current strategy in the search for life on Mars is to "follow the water" these areas are of primary interest for conducting additional in situ investigations.

A vehicle with the ability to traverse across and around the gullies is needed to conduct an in-situ investigation. While this is currently not feasible using conventional rovers, a new unconventional vehicle known as a Tumbleweed rover could potentially be used. Designed to derive mobility through use of the surface winds on Mars, Tumbleweed rovers would be lightweight and relatively inexpensive, allowing multiple rovers to be deployed in a single mission to particular areas of interest. Tumbleweeds would be used to complement currently planned missions by conducting surveys, pinpointing locations of interest for detailed follow-on investigations by rovers, landers, or perhaps human explorers. NASA Langley Research Center (LaRC), is currently studying deployable structure Tumbleweed concepts.

A mission would begin by deploying a group of Tumbleweed rovers on the upslope plateau behind an area with gullies. The Tumbleweeds would first characterize the plateau, which would include subsurface sounding to detect an aquifer if it exists. The Tumbleweeds, equipped with a simple ability to stop and start (e.g., changing shape of the structure), would then proceed toward the gullies when the wind direction is favourable. Proceeding down the slope, the Tumbleweeds would continue the search for evidence of a shallow aquifer, which would take the form of an ice plug beneath the overlying plateau, but close to the cliff face surface. Evidence of liquid water would also be searched for on the exposed portions of the rock layers. As the Tumbleweeds proceed down the channels, the interior of the channels would be examined to provide improved measurements regarding channel bed shape, channel depth, channel path (i.e., deflection around obstacles), etc. The Tumbleweeds would complete their mission by examining the debris apron to determine size of particles being transported down-slope and the composition of the soil in the debris apron.

Many challenges exist for implementing this mission using a Tumbleweed rover. For example, how can the aerodynamic properties of a Tumbleweed rover be "controlled" in order provide a stop/start capability that will allow the vehicle to not only make measurements at particular locations but also allow it to wait for favourable wind conditions? What are the wind conditions near Martian gully regions? This paper will address these issues and provide an in-depth discussion on the latest scientific findings concerning the gullies and their potential formation mechanisms.

The Mars Astrobiology Probe: A proposed instrument suite for the 2009 Mars Science Laboratory (MSL)

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We have proposed for the MSL Investigation to carry out a series of experiments using a groundbreaking new instrument concept, the Mars Astrobiology Probe (MAP). The MAP experiments are designed to address in a robust and comprehensive manner one of the primary goals of the MSL investigation, which is "to assess the biological potential of a least one target environment identified prior to MSL or discovered by MSL". MAP consists of four separate major components: a subcritical water extractor (SCWE); the Mars Organic Detector (MOD): a novel lab-on-a-chip micro-capillary electrophoresis (CE) system; and the Mars Organic Reactor Suite (MORS). SCWE is used to extract the target compounds from samples provided by the MSL sample distribution system. MOD next uses sublimation at Mars ambient pressure to extract, purify and concentrate organic compounds from the SCWE extract. MOD then assesses presence of two target classes of compounds, amines/amino acids and polycyclic aromatic hydrocarbons (PAHs), by measuring the fluorescent response on a MOD capture cold finger. In the case of amino acids, fluorescence is generated by their reaction with a dye, fluroescamine, which is highly specific for primary amines. PAHs are naturally highly fluorescent so they can be detected directly. To permit the simultaneous detection of both target compounds, half of the cold finger is coated with the fluroescamine reagent, while the other half is uncoated. With the MOD fluorescence analyzer, the target compounds can be readily detected at the sub-ppb level. If either amines/amino acids or PAHs or both are detected in the MOD-based screening of a sample this means that other organic compounds are also likely present in the sublimate. Thus, other organic detection instruments selected for MSL may want to interrogate the sublimed material to characterize these compounds. If an amine/amino acid signal is detected in the MOD-based analyses, the microfabricated chip-based CE separation device with integrated reaction chambers, pumps, and capillary sipper is used to gather the sublimate from the fluroescamine coated portion of the MOD cold finger and determine the amino acid composition and chirality in order to evaluate their origin. Of particular importance would be the finding that the amino acids are present as a non-racemic mixture (non-equal amounts of the D- and L-isomers), which could be suggestive of a biotic origin. The SCWE/MOD/CE suite combines with the MORS component that is used to determine the oxidative characteristics of the samples in order to provide data on the role of oxidation reactions in the survival of organic compounds in the Martian regolith. These oxidant data will be of particular significance if a negative result (no fluorescent signal above background is detected) is obtained by the MOD. MAP has the potential of performing the first successful detection of organic compounds on Mars. The finding of amino acids of possible biological origin would be a sensational result of interest to both the scientific community and the public. The MAP project also includes an extensive education and public outreach effort in order to transmit the results on a national and worldwide basis.

Mars Astrobiology Science and Technology Workshop, 8-10 September 2004, Carnegie Institution of Washington, Washington, DC http://astrobiotech.arc.nasa.gov/abstract_deadline.cfm

THE ULTRASONIC/SONIC DRILLER/CORER (USDC) AS A SUBSURFACE DRILL, SAMPLER AND LAB-ON-A-DRILL FOR THE MARS ASTROBIOLOGY SCIENCE AND TECHNOLOGY

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The search for existing or past life in the Universe is one of the most important objectives of NASA's mission. For this purpose, effective instruments that can sample and conduct in-situ astrobiology analysis are being sought. In support of this objective, a novel Ultrasonic/Sonic Driller/Corer (USDC) based mechanism has been developed to probe and sample rocks, ice and soil. The USDC consists of an ultrasonic actuator that impacts a coring or drilling bit at sonic frequencies through the use of an intermediate free mass. The USDC can produce both a core and powdered cuttings as well as emit elastic waves. For planetary exploration, this mechanism has the important advantage of requiring low axial force, virtually no torque, and can be duty cycled to require as low power as 2-W. This low axial load advantage overcomes a major limitation of planetary sampling in low gravity environments and when operating from lightweight robots and rovers. The low power operation produces a minimum temperature rise which is required for the acquisition of biologically meaningful samples.

The development of the USDC is being pursued on various fronts ranging from analytical modeling to mechanisms improvements while seeking a wide range of applications. While developing the analytical capability to predict and optimize its performance, efforts are made to enhance its capability to drill at higher power, high speed while operating quietly. Taking advantage of the fact that the bit does not turn and that it is only subjected to minute displacements, sensors (e.g., thermocouple and fiberoptics) were integrated into the bit to examine the borehole during drilling. The sounding effect of the drill was used to emit elastic waves in order to evaluate the surface characteristics of rocks. As a corer, samples were made from various rocks, including basalt and limestone, at dimensions that are as large in diameter as 5-cm and as long as 10-cm. Studies conducted in conjunction with CHEMIN (Chemical and Mineralogy analysis) task, showed that the produced micron size powdered cuttings had dimensions that are ideal for X-Ray Diffraction (XRD) analysis. To take advantage of the low axial load requirement of the USDC, a 4-legged walking robot is currently being developed to climb steep rocks using a USDC on each of the legs to anchor the rover to the rocks while climbing. Upon completion of this task, the possibility of walking on concrete ceiling will be considered. For deep surface penetration, a U/S gopher that is 5-cm in diameter and about 1-m long is being developed to reach as deep as 20-m at -20^{o}C ice in Lake Vida, Antarctica. The scaling of the USDC is also an issue that is being investigated, where besides the U/S gopher we are considering the development of a U/S jackhammer. In this paper, the latest status of the USDC development and applications that are underway will be reviewed.

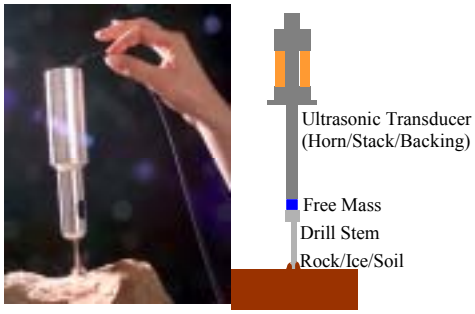

FIGURE 1: The USDC is shown coring with minimum axial force and holding torque (left), and a schematic diagram of the USDC components (right).

Using protein-DNA chimeras to detect and count small numbers of molecules

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Quantification of specific molecules is central to many fields, including biology, where differences in the number of low abundance proteins can bring about profoundly different cellular consequences. We describe general methods to detect and quantify small numbers of specific molecules. We redirected self-splicing protein inteins to create "tadpoles", chimeric molecules comprised of a protein head covalently coupled to an oligonucleotide tail. We made different classes of tadpoles that bind specific targets, including *Bacillus anthracis* protective antigen and the enzyme cofactor biotin. We measured bound target by quantifying DNA tails by T7 RNA polymerase runoff transcription and by real-time PCR. Assays based on these reagents had dynamic ranges of more than 11 orders of magnitude and distinguished numbers of molecules different by as little as 10%. At their low limits, they detected as few as 6400 protective antigen molecules, 600 biotin molecules, and 150 biotinylated protein molecules. In crudely fractionated human serum, they detected as few as 32,000 protective antigen molecules. Tapdoles enable simple and sensitive measurements of the number of molecules. By hybridization of their nucleic acid tails, they may open paths to new materials and nanostructures.

TECHNICAL APPROACHES TO LASER MASS SPECTROMETRY AT MARS.

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Overview. Laser mass spectrometry may be a key element of future astrobiological investigations at Mars. A focused laser can evaporate sample from small spots (microns to mm) which may be selected with contextual data from a microscope and other probes. The point-by-point elemental and molecular composition obtained through subsequent mass spectrometry is vital to (i) determine the spatial associations of potential biosignatures with structures and/or mineral phases, and (ii) detect high molecular weight compounds that may be highly localized. The broad-band analysis of complex samples by laser mass spectrometry is well-documented, but for implementation at Mars a number of key technical areas have required special attention.

Sample Processing. Laser desorption (LD) has the advantage of sampling an unacquired/unprepared surface from a stand-off position. While this suggests a highly simple implementation on airless bodies, the thick atmosphere on Mars necessitates additional considerations and approaches. It is possible to perform laser desorption at ambient pressure and draw neutrals/ions into an evacuated mass spectrometer using only a pressure gradient [1]. The advantage is that no sample acquisition would be required. The challenge is that the inlet must be placed very close (mm-scale) to the surface to achieve good sensitivity. Alternately, samples are acquired and placed in vacuum through a load lock. This approach permits precise manipulation and processing of the sample, if required, and achieves high sensitivity, but does require a robust, repeatable mechanical sealing system [2]. Hybrid approaches, such as acquiring and positioning samples at ambient pressure, followed by laser desorption, are also being considered.

Pulsed Lasers. Pulsed LD, such as using diode-pumped or semiconductor YAG-based systems with a Q switch, comprise an enormously powerful enabling technology for *in situ* astrobiology. Short pulse lengths (ps to ns scale) enable the volatilization and ionization of high-mass "parent" compounds, from geological samples, that would be uniquely diagnostic of the current organic complexity of Mars; this is difficult to do with non-laser methods. Relatively simple "direct" LD can evaporate and ionize with a single laser shot. Multi-laser methods such as resonance ionization offer improved sensitivity and selectivity at increased complexity. Because flight-qualifying a particular system is a complex and expensive task, brassboard work on alternate approaches is being conducted in the laboratory to determine the most effective route for Mars.

Mass Analyzers and Detectors. The applicability of various analyzer types (time-of-flight, ion trap, quadrupole, etc.) to specific analytical challenges on Mars is being investigated. Each has its advantages (high mass, tandem capability, sensitivity, etc.) and the best approach is likely a carefully-tailored and streamlined combination of these [2]. Quite miniature analyzers have been developed in recent years, and space implementation may now be equally dependent on key supporting technologies such as vacuum pumps and electronics. For instance, a single-board, low-power, rad-tolerant, high-bandwidth waveform digitizer, under development at JHU/APL, is required to resolve and integrate the tightly-spaced peaks of a time-of-flight mass spectrum.

Acknowledgement. This work has been supported by NASA ASTID and MIDP grants.

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High Resolution Electron-Induced XUV Fluorescence for Life Detection

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The most direct evidence for life on other planets would be to detect organic material, but this can be destroyed by environmental degradation within a few tens of millions of years. In contrast to organic substances, mineral fossil evidence can survive for several billion years. Our goal is, therefore, to determine how to recognize the mineralogical traces of life that could be in extraterrestrial rocks.

The Viking and Pathfinder missions have shown that Fe and Mn exist in a number of oxidation states on the Martian surface, for example, FeO, Fe_2O_3, Fe_3O_4, MnO, MnO_2, or Mn_2O_3. Minerals formed by bacteria that metabolise Fe and Mn (by redox processes) in such molecules differ from similar minerals formed through chemical weathering. In particular, a steeper gradient in the spatial distribution of the charge states of the constituent ions Fe^{q+} or Mn^{q+} (q = 1, 2, …) than typical of a non-biological mineral would suggest previous biological processes. The energy dispersive hard X-ray fluorescence technique, for example, has been widely used to determine elemental composition, but it cannot distinguish between the different charge states Fe^{q+} or Mn^{q+}. We have therefore developed a soft X-ray (XUV) emission experiment to determine the spatial distribution of the oxidation states, which thus makes possible the search for new evidence of life.

In the experiment, we bombard the sample with energetic electrons (1-20 keV) and observe the resulting K or L shell fine structure emission lines. The energies of the fine structure lines depend on the charge of the metal ions Fe^{q+} or Mn^{q+} in the molecule emitting the radiation. It is therefore possible to identify the oxidation state of a mineral by measuring the energies of the K and L shell lines. For example, we show in Fig. 1 the energy shifts of the $L_{2,3}$ ($3d \rightarrow 2p_{1/2,3/2}$) emission lines for different oxidation states of Fe in some background minerals on Mars, where the data was collected at the Lawrence Berkeley Laboratory. We have also found that the oxygen KL emission line ($2p \rightarrow 1s$) of a biological sample (not shown) is narrower than the same line in non-biological minerals. The soft X-ray (XUV) region is thus important in the search for life.

The method is suitable to examine planetary materials returned to Earth from Mars and could be miniaturized to scan samples *in situ* on the planetary surface.

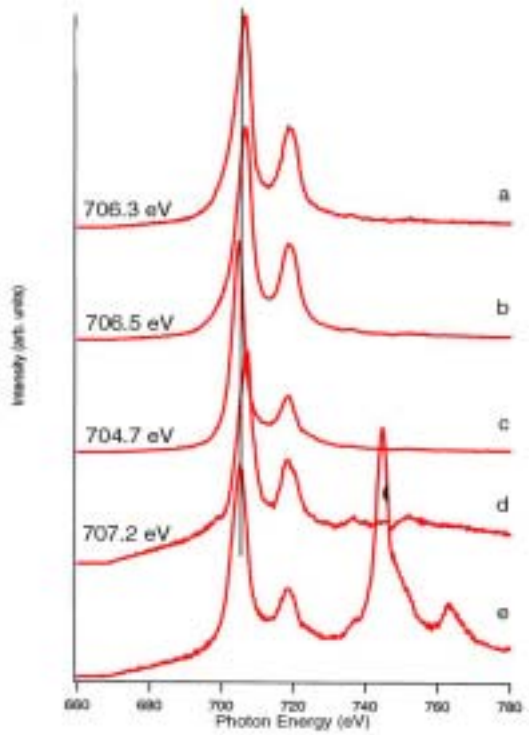

Fig. 1. The $L_{2,3}$ lines of a) $Fe^{3+}{}_2O_3$ (Hematite), b) $Fe^{3+}{}_2Fe^{2+}O_4$ (Magnetite), c) Fe, d) $Mg_{1.6}Fe^{2+}{}_{0.4}(SiO_4)$ (Olivine) and e) Fe and Ni. The energies of the L_3 lines are shown.

SOLVENT EXTRACTION AND CHEMICAL DERIVATIZATION OF ORGANIC MOLECULES OF EXOBIOLOGICAL INTEREST FOR IN SITU ANALYSIS OF THE MARTIAN REGOLITH

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Mars is presently the most likely planet on which there is a possibility of finding extinct and/or extant life. Future exploratory missions to Mars in search of evidence for life will focus on key organic molecules such as carboxylic and amino acids. The 2009 Mars Science Laboratory (MSL) Mission will offer an opportunity to carry out in situ measurements for organic compounds on Mars. , Gas chromatography mass spectrometry (GC/MS) is one technique that will be proposed for MSL. We are currently developing an automated extraction process coupled to chemical derivatization in order to target several key organic compounds using GCMS.

This paper presents a solid-liquid extraction method (1) that can be coupled with in situ GC/MS analyses of organic compounds on Mars. The extraction efficiencies of several different organic solvents including (isopropanol and water) have been determined ,for several Martian soil analog materials such as an Atacama Desert soil sample from Chile (2). It was shown that isopropanol is the best solvent, allowing high extraction yields for both amino and carboxylic acids with space compatible extraction time (15 to 30 min) when the extraction procedure is assisted by sonication. A highly sensititive and quantitative single-step derivatization reaction was carried out using N-methyl, N-tert.-butyl (dimethylsilyl) trifluoroacetamide (MTBSTFA) as the silylating agent prior to GC/MS analysis. The development of a miniaturized reactor, where both the extraction and the derivatization processes could take place is currently under investigation. This method is discussed for an easy automation with coupling to an in situ GC-MS space instrument.

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(2) R. Navarro-Gonzàlez et al., *Science* 302 (2003) 1018-1021

Measurement of Microbial Activity in Soil or Atmospheric Dust by Colorimetric Observation of *in situ* Dye Reduction: an Approach to Detection of Extraterrestrial Life

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Abstract

Detecting microbial life in extraterrestrial locations is a goal of space exploration because of ecological and health concerns about possible contamination of other planets with earthly organisms, and *vice versa*. Previously we suggested a method for life detection based on the fact that living entities require a continual input of energy accessed through coupled oxidations and reductions (an electron transport chain). We demonstrated using earthly soils that the identification of extracted components of electron transport chains is useful for remote detection of a chemical signature of life. More recently we used Earth-derived soils to develop a related, but simplified life detection system based on direct observation of a biological redox signature. We measured the ability of soil microbial communities to reduce the artificial electron acceptors 2,3-dichlorophenol indophenol (DCIP) and the tetrazolium dye 2,3-bis(2-methoxy-4-nitro-5-sulfophenyl)-2H-tetrazolium-5-carboxanilide inner salt (XTT). Uninoculated or killed controls did not reduce the dyes. A soil from Antarctica that was determined by chemical signature and DNA analysis to be sterile also did not reduce the dyes. The present version of the technology employs a robotic instrument designed to collect dust or aerosol particles from air and deposit these materials into a bank of nutrient solutions containing various microbial growth media and DCIP or XTT. A remote camera monitors color changes resulting from microbial metabolism in the media that cause dye reduction (color formation with XTT; color quenching with DCIP). Observation of dye reduction provides a simplified means to detect a signature of life in the soils or atmospheric dusts of other planets or their moons.

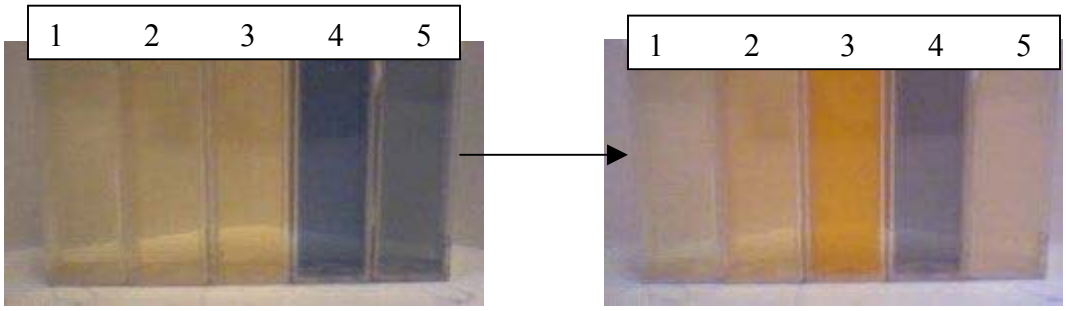

Figure 1: Robotic examination of dust or aerosol samples for living microorganisms

1 = Killed Control (no dye): negative; 2 = Killed Control (XTT): negative; 3 = Positive (XTT) kan^r amp^r *E. coli*: **positive;** 4 = Killed Control (DCIP): negative; 5 = Positive (DCIP) kan^r amp^r *E. coli*: **positive;** Killed controls contained ~1% *p*-cresol; 7-hour observation at 25°C.

Molecular Recognition Sensing Materials within Operational and Mission Environments for Planetary Exploration: Consideration of Stability and Robustness.

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The question of life elsewhere in the Universe is central to the discipline of astrobiology. *In situ* exploration for past or present life in candidate locations within the Solar System is envisaged via a number of proposed missions to locations such as Mars and Europa.

An approach to identify life is via (i) detection of molecular evidence of past or present life in the form of organic molecular biomarkers together with (ii) an understanding of the contextual abiotic organic environment in which biomarkers may be found. The latter is also of interest in the exploration of other bodies such as comets and asteroids.

For environmental and medical applications on Earth that desire *in situ* detection and characterisation of trace organic markers, molecular receptor based sensor technologies are receiving much attention. The current trends of multi-analyte sensor arrays, μ-fluidic and μ-system approaches, and transducer readout technology development is making remote analytical systems for astrobiology and planetary exploration a realistic practicality. A number of instrument concepts for planetary exploration have been proposed including MASSE, MoBiLD and SMILE.

Such "biosensor" technologies rely fundamentally upon the integration of molecular receptors into devices to recognise and bind complementary organic molecular targets of interest. Such sensors can exploit a wide range of molecular recognition materials with key examples being (i) proteinaceous antibodies and their various recombinant derivatives and (ii) artificial molecular recognition materials such as wholly synthetic molecular imprinted polymers (MIPs).

The operational and mission environments for an anticipated Martian mission offer a considerable challenge for the use of "fragile" molecular recognition materials due to the diversity of extreme environments that are encountered. The following summarise the key components: (i) acoustic energy – *e.g.* during launch, (ii) sterilisation – *e.g.* gamma radiation, thermal baking, chemical, H_2O_2 plasma, *etc*., (iii) thermal – both cycling and extreme excursions, (iv) extended storage times including pre-launch, (v) radiation – both particle and gamma radiation from Solar and cosmic sources and (vi) chemically aggressive sample matrices.

For many of the preceding environments, little testing of molecular recognition materials has been previously performed. This presents a significant deficiency in the current knowledge required to allow appropriate early systems level consideration of future missions.

We have initiated a series of studies to explore the stability of molecular recognition materials in a variety of operational and mission simulations and will report on the latest findings.

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Unit Operations & Application of Microfluidic Systems for Remote Analysis

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Within the last decade the concepts of miniaturization have been seriously applied to chemical and biological problems. Of particular focus and interest has been the development and application of microfluidic or 'lab-on-a-chip technology'. These microscale analytical instruments employ micromachined features (such as channels, electrodes, reactors, and filters) and are able to manipulate fluid samples with high precision and efficiency. Microfluidic systems have been used in a wide variety of applications including nucleic acid separations, protein analysis, process control, small-molecule organic synthesis, DNA amplification, immunoassays, DNA sequencing, and cell manipulations. In a fundamental sense, chip-based analytical systems have been shown to have many advantages over their conventional (larger) analogues. These include improved efficiency with regard to sample size, response times, cost, analytical performance, process control, integration, throughput, and automation [1].

Much of the pioneering work in microfluidics has focused on the successful transfer of established analytical technologies from conventional to microfluidic (chip-based) formats. In particular, huge leaps in the efficiency and application of separation techniques have been facilitated by miniaturizing column dimensions and creating monolithic fluidic networks on planar substrates. Furthermore there is much current interest in using microfluidic systems for chemical and biological synthesis. Because of the unique environments afforded within microfluidic networks, a variety of synthetic processes can be performed in continuous flow and batch formats. Increased efficiencies of mixing and separation combined with high rates of thermal and mass transfer make microreactors ideal for processing valuable reaction components and improving reaction selectivities [2].

The idea that microfabricated analysis systems could be used in extraterrestrial environments is not new. The small size and low power requirements of the first silicon gas chromatograph fabricated by Stephen Terry at Stanford University in 1975 were seen at the time as ideal characteristics for better utilizing spacecraft resources. At a fundamental level, chip-based analysis systems possess many distinct advantages over their conventional counterparts for space flight purposes. These include improved performance, reduced instrumental footprints, reduced system masses, low power requirements, high levels of functional integration, built in redundancy (i.e. multiple devices per monolith), lack of moving parts and operational flexibility [3]

The paper will describe how we have used microfluidic devices to perform key unit operations within monolithic chip-based systems. Particular emphasis will be placed on component processes likely to be necessary for operation in extraterrestrial environments. These include sample pre-concentration [4], biomarker separation and highly miniaturized and integrated detection technologies [5]. It is hoped that the case studies presented will provide strong arguments for the use of microfluidic systems in future space missions.

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Optical Sensors for Biomolecules Using Nanoporous Sol-Gel Materials

by

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An important consideration for space missions to Mars is the ability to detect biosignatures. Solid-state sensing elements for optical detection of biological entities are possible using sol-gel based biologically active materials. We have used these materials as optical sensing elements in a variety of bioassays, including immunoassays and enzyme assays. The sol-gel approach produces an optically transparent 3D silica matrix that forms around the biomolecule of interest, thus stabilizing its structure and functionality while allowing for optical detection. This encapsulation process protects the biomolecule and leads to a more "rugged" sensor. The nanoporous structure of the sol-gel matrix allows diffusion of small target molecules but keeps larger, biomolecules immobilized in the pores. By immobilizing an appropriate biomolecule in the sol-gel sensing element, we have successfully detected analytes such as amino acids and hormones. In the case of the amino acid glutamate, the enzyme glutamate dehydrogenase was the immobilized molecule, whereas in the case of the hormone cortisol, an anti-cortisol antibody was immobilized in the sensing element. In this previous work with immobilized enzymes and antibodies, excellent sensitivity and specificity were demonstrated in a variety of formats including bulk materials, thin films and fibers. We believe that the sol-gel approach is an attractive platform for bioastronautics sensing applications because of the ability to detect a wide range of entities such as amino acids, fatty acids, hopanes, porphyrins, etc.

ASTROBIOLOGICAL APPLICATIONS OF THE MBARI ENVIRONMENTAL SAMPLE PROCESSOR (ESP). S. M. Feldman[1], C. Scholin[2], J. Feldman[2], S. Jensen[2], B. Roman[2], C. Preston[2], V. Orphan[3], and J. Dzenitis[4] [1]NASA Ames Research Center, MS 239-4, Moffett Field, CA 94035 (Sabrina.M.Feldman@nasa.gov), [2]Monterey Bay Aquarium Research Institute, [3]California Institute of Technology, [4]Lawrence Livermore National Laboratory.

The Monterey Bay Aquarium Research Institute (MBARI) has developed an Environmental Sample Processor (ESP) for autonomously sampling and detecting microbes found in terrestrial ocean environments. This instrument can readily be adapted for exploring deep-sea seep and hydrothermal vent fluids as one step towards developing the scientific and technical capability for searching for life on other planets. With moderate modifications, the ESP could also be adapted for Martian surface applications including detection of astrobiologically relevant prebiotic and biotic compounds that may exist in dry, icy, or liquid samples.

The ESP provides an autonomous platform for sample acquisition, processing, and distribution, and enables a variety of molecular probe chemistries as well as options for sample archival. Furthermore, the architecture of the ESP permits expansion of analytical functions that share a common requirement for front-end sample collection and processing prior to delivery to a particular analyte detector.

The first generation ESP has already been demonstrated in the field and a second generation is currently under development in the laboratory. The ESP consists of five major subsystems: carousel, shuttle, clamp, syringe pump, and CCD camera (Fig. 1). The carousel stores ~100 "pucks", or reaction chambers, which accommodate a wide variety of user-defined 25 mm diameter filters or chemically adsorptive media. An elevator and linear shuttle are used to move a puck from the carousel to the processing position where it is sealed in a clamp, thus providing connections to the sample port and reagent valve manifolds. The seals used in the clamp have embedded heater pads for temperature control from ambient to ~100°C at any time during a protocol. The shuttle is also used to move pucks to an imaging station where a CCD camera records results of DNA probe array assays. A syringe pump draws in seawater samples and dispenses the required reagents. Modular valving supports use of up to 16 different custom-defined reagents.

Figure 1. Solid model of the second generation ESP. The rotating carousel, CCD camera, puck clamps, syringe pumps, valving and reagent bags are visible. The ESP is protected under US Patent No 6187530.

In order to illustrate the capabilities of the ESP, we will present results from the first generation instrument and describe modifications to the ESP which could allow it to be used for (1) evaluating the diversity and abundance of thermophilic and methanotrophic microorganisms present in deep-sea hydrothermal vents; and (2) detecting a variety of organic compounds on the Martian surface.

ASTROBIOLOGICAL SIGNIFICANCE OF DEFINITIVE MINERALOGICAL ANALYSIS OF MARTIAN SURFACE SAMPLES USING THE CHEMIN XRD/XRF INSTRUMENT. S. M. Feldman[1], D.F. Blake[1], P. Sarrazin[1], D. L. Bish[2], S. J. Chipera[3], D. T. Vaniman[3], and S. Collins[4] [1]NASA Ames Research Center, MS 239-4, Moffett Field, CA 94035 (Sabrina.M.Feldman@nasa.gov), [2]Dept. Geological Sciences, Indiana University, Bloomington IN 47405, [3]Hydrology, Geochemistry, and Geology, Los Alamos National Laboratory, MS D469, Los Alamos, NM 87545, [4]CCD Imaging Group, Jet Propulsion Laboratory, MS 300-315L, Pasadena, CA 91109

The search for evidence of *habitability*, or of extant or extinct life on Mars, will initially be a search for evidence of past or present conditions supportive of life. The three key requirements for the emergence of life are thought to be liquid water; a suitable energy source; and chemical building blocks. CheMin is a miniaturized XRD/XRF (X-Ray diffraction / X-ray fluorescence) instrument which has been developed for definitive mineralogic analysis of soils and rocks on the Martian surface [1, 2]. The CheMin instrument can provide information that is highly relevant to each of these habitability requirements as summarized below.

Liquid water. CheMin can easily identify any minerals which require liquid water for their formation as well as mineralogical alteration products caused by the presence of liquid water.

Chemical Energy Sources. The CheMin instrument can also provide information on mineralogical energy sources for chemosynthetic lifeforms such as manganese(II) which can be oxidized to manganese(IV), ferrous iron which can be oxidized to ferric iron, and sulfide which can be oxidized to sulfate. In addition, CheMin can identify geochemical reactions such as the conversion of olivine to serpentine (which occurs only in the presence of liquid water) which could serve as an energy source for microbial organisms through the production of H_2 (the basis for many chemoautotrophic biochemical processes).

Chemical Building Blocks. The CheMin instrument will provide information on both the major elements (H, C, O, N, S, and P) and minor/trace elements thought to be required for life. These elements can be indirectly detected (e.g. through the identification of carbonates, phosphates, sulfates, and nitrates) or directly detected by the XRF spectrometer for $8 < Z < 92$. The CheMin instrument can also identify mineralized organic materials and biomineralization products.

Other habitability considerations. Definitive mineralogical analyses of Martian rock and soil samples can establish the environments of formation and alteration of a rock sample by determining temperature and pressure conditions of formation, present/past climate, water activity, the fugacity (activity) of biologically significant gases and the like. This information could provide insight into the early history of Martian volatiles and potentially establish the presence and lateral extent of hydrothermal systems.

In order to illustrate the CheMin instrument's capabilities, we will present results from the CheMin-III instrument (developed to the TRL-6 level) and describe three example Martian surface investigations in which CheMin data could be used to evaluate evidence of present or past habitability. The three are: (i) Evaporite rock strata such as found by MER-B at Meridiani Planum; (ii) an impact-gardened basalt environment such as found by MER-A at Gusev Crater; and (iii) a global Mars soil unit.

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MEMS-based Force-Detected Nuclear Magnetic Resonance Spectrometer system for Astrobiological Investigations

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The proof-of-concept for a highly-miniaturized, Force-Detected Nuclear Magnetic Resonance (FDNMR) Spectrometer integrated with microfluidic sample extraction and handling is being developed under ASTID funding. Nuclear Magnetic Resonance (NMR) is the premier spectroscopic technique for the identification of organic compounds in solution. NMR spectrometers are ubiquitous in terrestrial chemical laboratories and are used extensively for elucidating the chemical structure, composition and bonding dynamics of organic molecules. Although attempts have been made in the past to miniaturize conventional NMR spectrometers, these approaches suffer from the very narrow range of molecular species that can be studied and the relatively large quantities of sample that are required. The proposed 2-mm diameter FDNMR spectrometer will be the world's smallest NMR detector capable of studying 60-μm-sized liquid samples, with superior sensitivity at these sample sizes in comparison to the conventional, inductive detection approach. It is envisaged that once the proof-of-principle for such a system has been demonstrated, it will be possible to develop "massively parallel" detection schemes involving several spectrometers mounted on a single microfluidics transport system, providing high-throughput, multiple analyses.

The development of an integrated FDNMR system will be undertaken over a period of three years. The first year effort is devoted primarily to the separate fabrication, assembly and testing of a MEMS FDNMR spectrometer and a micro-capillary based microfluidics sample transport system. We will report preliminary research results obtained from this work.

CARBON AND SILICON NANOWIRE CHEMICAL SENSORS FOR IN SITU ASTROBIOLOGICAL MEASUREMENTS*. Brian Hunt, Mike Bronikowski, Anita Fisher, and Eric Wong, NASA/JPL/Caltech, Pasadena, CA 91109, bdhunt@jpl.nasa.gov.

Nanowire-based chemical sensors utilize a simple and effective sensing mechanism based on the change in conductance of a semiconducting nanowire (NW) as a molecule adsorbs on the surface of the wire. These devices act as chemically sensitive field-effect transistors (FETs) in which conductance changes arise from electrostatic gating or charge transfer from attached molecules. Such sensors have already demonstrated detection limits in the picomolar to femptomolar range for a few model molecular systems [Hahm and Lieber, Nano Letters **4**, 51 (2004)]. The nanowire molecular sensors typically operate at sub-nW power levels and can be made chemically specific by functionalization of the nanowire surface, as illustrated in Figure 1.

Here we report on our work on nanowire chemical sensors utilizing carbon nanotubes (CNT) and silicon nanowires grown by chemical vapor deposition (CVD) from nanoscale catalyst particles. The CNT devices are produced using lithographically patterned molybdenum electrodes followed by definition of an iron nanoparticle catalyst region and CVD growth of the nanotubes, resulting in single-wall nanotubes spanning electrode gaps ranging from 100 nm to 2 microns (Figure 2). The Si NW are grown from Au nanoparticle catalysts or nm-thickness Au films, followed by patterning of Ti/Au electrodes. To date, our chemical sensing efforts have focused on bare, unfunctionalized nanowires, primarily CNTs. We have demonstrated both gas and liquid phase chemical sensing using the CNT devices. The liquid phase testing has been aimed at measurements of amino acids in water, as key molecules of interest in astrobiology. Preliminary conductance-versus-time measurements of electrically gated nanotube sensors were made as the concentrations of three different amino acid solutions were varied. These experiments showed clear differences in the conductance change for arginine, aspartic acid, and tryptophan. Further studies are underway to optimize the sensitivity via adjustment of the device gate voltage and to understand the effect of pH. These initial measurements suggest that even unfunctionalized nanotubes may be useful for amino acid sensing. We will also report on our ongoing efforts to functionalize Si/SiO_2 and CNT surfaces to enable chemically specific detection of biomolecules of interest. The initial functionalization studies have utilized oxysilane chemical linkers on Si/SiO_2 to attach biotin as a model receptor. Chemically specific reaction with the model target molecule, streptavidin labeled with Au nanoparticles, was verified by AFM and SEM studies. Such functionalization may eventually enable chemically specific sensing of a variety of different molecules in nanowire sensor arrays, as well as chirally specific sensing of biomolecules as a potential indicator of extraterrestrial life. **Work supported by NASA Code R/T BioNano program.*

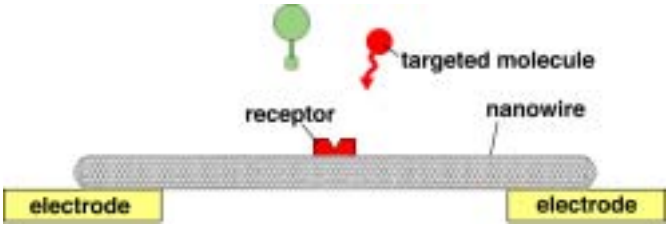

Figure 1. Schematic illustration of chemical sensor using functionalized CNT and conductance modulation.

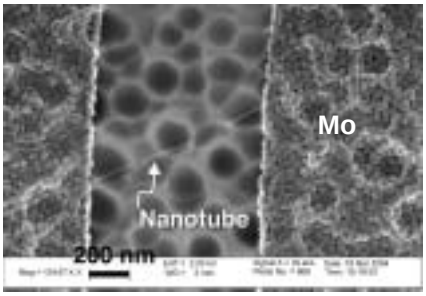

Figure 2. SEM picture of CNT bridging gap between Mo electrodes.

WET CHEMISTRY EXPERIMENT AT MARS (WETCHEM)

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NASA is currently developing strategies for several in-situ missions to Mars during the coming decades to explore and quantitatively assess the potential habitability of the Martian surface. Some of these missions will be designed to specifically search for biosignatures. In order to analyze inorganic and organic compounds from surface and sub-surface samples, and to search for biosignatures, we propose a novel experimental approach utilizing wet chemistry combined with mass spectroscopy called "Wet Chemistry Experiment at Mars (WetChem)". WetChem consists of an integrated suite of miniature instruments: a Robotic Chemical Analysis Laboratory (RCAL), and an Electrospray Ionization/Ion Mobility spectrometer (ESI/IMS) in tandem with a Cylindrical Ion Trap Mass Spectrometer (CIT-MS). Fig.1 summarizes WetChem's science and measurement objectives.

RCAL is designed to provide information about the aqueous chemistry and soluble minerals present in the Martian soil using an array of electrochemically-based sensors. The sensor array consists mainly of potentiometric ion selective electrodes but also includes conductivity, and voltammetric microelectrodes for determination of heavy metals. In addition, RCAL will serve as the front-end instrument to extract volatiles, inorganic and organic compounds from the Martian regolith and feed them to the ESI unit for further analysis by IMS/CIT-MS. Via the MS we will be able to conduct a survey for a broad variety of organic molecules, enabling the identification of potential chemical biosignatures (i.e. amino acids, carboxylic acid). Mass spectrometric methods will also be used for chiral amino acid analysis.

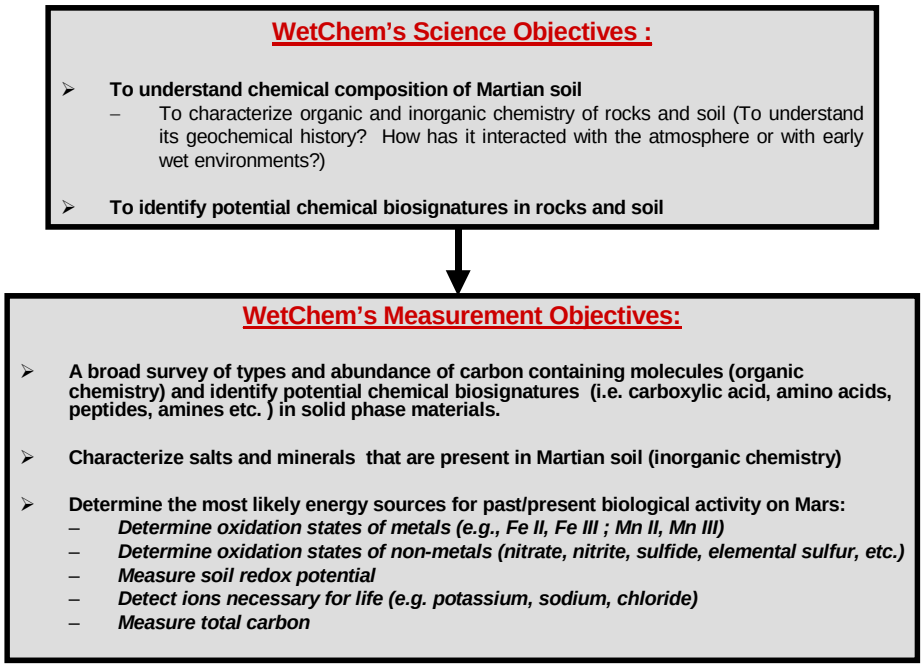

Fig. 1 Summary of WetChem's science and measurement objectives.

THE 2007 PHOENIX MARS SCOUT MECA WET CHEMISTRY LABORATORY

S. P. Kounaves[1], M. H. Hecht[2], P. Smith[3], and The Phoenix Team

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The MECA *Wet Chemistry Lab* (WCL), originally developed as part of the *Mars Environmental Compatibility Assessment* (MECA) package for the since cancelled 2001 Mars Surveyor Program lander, is one of the instruments included in the upcoming 2007 Phoenix Mars Scout mission. In addition to investigating the geochemistry, the MECA-WCL directly addresses a variety of astrobiology goals. Among these is its search for habitable zones and biosignatures by, (1) identifying potential chemical energy sources available to support life, (2) determining whether the subsurface geochemistry is hostile to life, and (3) identifying the potential of the geochemical environment to preserve paleontological evidence.

Its capability to achieve astrobiologically oriented goals is provided by an array of electrochemically-based sensors. The array consists of *ion selective electrodes* (ISE) for inorganic anions, cations, and gases, including calcium, sodium, potassium, magnesium, chloride, bromide, nitrate, perchlorate, carbonate, dissolved CO_2 , O_2, and pH. It also includes special electrodes for conductivity, oxidation - reduction potential (ORP), *anodic stripping voltammetry* (ASV) for heavy metals such as Cu^{2+}, Cd^{2+}, Pb^{2+}, Hg^{2+}, and *cyclic voltammetry* (CV) for identifying and analyzing possible reversible and irreversible redox couples.

The 2007 Phoenix lander includes four WCLs, each consisting of an upper sample/water/reagent *dispensing unit* with a stirrer and a lower *analysis cell* (beaker) containing the integral sensor array. Each WCL contains a drawer which can accept a soil sample from the robotic arm and deliver approx. 1 cc of soil sample to 25 cc of water containing a low concentration of inorganic ions as a standard and also acting as a leaching solution. In addition to being flight qualified for the MSP'01 mission, the WCL has been rigorously and successfully tested after being frozen for over 18 months, thawed and refrozen dozens of times, and after sitting on the shelf for over a year at room temperature. The figure at he right shows the upper (dispenser) and lower (beaker) assemblies.

The WCL for the 2007 Phoenix, and future Mars missions, provides a low mass/energy device for obtaining unique information about the potential habitability and history of the aqueous and geochemical environment.

TUMBLEWEED: A WIND-PROPELLED SURVEY VEHICLE FOR ASTROBIOLOGY AND PLANETARY SCIENCE. K. R. Kuhlman[1], A. E. Behar[1], J. A. Jones[1], F. Carsey[1], M. Coleman[1], G. Bearman[1], M. Buehler[1], P. J. Boston[3], C. P. McKay[3], L. Rothschild[3], J. Antol[4], G. A. Hajos[4], W. C. Kelliher[4], I. A. Carlsberg[4], J. P. Keyes[4], M. Rudisill[4], R. L. Crawford[5], [1]Jet Propulsion Laboratory, California Institute of Technology, 4800 Oak Grove Dr., Pasadena, CA 91109 USA, kkuhlman@jpl.nasa.gov, [2]New Mexico Institute of Mining and Technology, 801 Leroy Place, Socorro, NM 87801 USA, [3] NASA Ames Research Center, Moffett Field, CA 94035 USA, [4]NASA Langley Research Center, Hampton, VA 23681-2199 USA, [5]Environmental Biotechnology Institute, Univ. of Idaho, Moscow, ID 83844-1052, USA

Introduction. Seasonally receding frost at Mars' higher latitudes provides liquid water; transient on the surface and longer-lived below it. Addition of sunlight and evaporitic dust offer possible surficial habitats, albeit only seasonally, although subsurface habitats might support life longer. Surficial habitats might populate a large area, perhaps from 50 degrees to the poles themselves, and this in turn suggests the significant value of a long-range survey for high latitude habitats. Site types particularly suitable to this survey include desert surfaces, duricrusts, pavements, ice surfaces and large flat-bottomed canyons. Such a survey could be readily accomplished with a fleet of Tumbleweeds - – large, inflatable, beach-ball-like vehicles capable of using the readily available wind to traverse the surface with minimal power, while optimizing their capabilities to perform a variety of measurements over relatively large swaths of terrain. Tumbleweeds have been proposed to conduct long-range, randomized surveys of habitability equivalent to conventional coordinate grid sampling. These vehicles will be released to roam for the duration of a season or longer, possibly on the residual martian ice cap.

Proposed Tumbleweed Deployments. Field campaigns in the Atacama Desert, Chile and the McMurdo Dry Valleys, Antarctica will demonstrate this approach on Earth and test the hypothesis: that water indicates habitability and microbial life will correlate with amount of water. Field campaign sites incorporating both snow surfaces and dry desert occur at high latitudes with more accessible hot deserts at mid to low latitudes. The Earth science objectives of these tests are to 1) document the heterogeneity of surface and near-surface habitability in these terrains and 2) measure the extent of biological activity within these habitats.

Instrumentation. The objectives of the proposed field campaigns will be accomplished by 1) outfitting the vehicle with mature instrumentation, 2) following the GPS trail of the Tumbleweed with a chase team equipped with a suite of field instrumentation and 3) analyzing samples collected along the route using a variety of laboratory instruments. The instruments onboard Tumbleweed include the Soil and Ice Conductivity Instrument (SICI), a commercial gas monitor capable of measuring oxygen, ammonia, carbon dioxide and hydrogen sulfide, a tunable diode laser (TDL) detector for water vapor, and a Computed Tomography Imaging Spectrometer (CTIS). These are the first set of instruments selected, but other low-mass, low-power instruments may be considered. The chase suite will include a portable gas chromatograph, ground penetrating radar and X-ray fluorescence (XRF) spectrometer. In-depth laboratory analyses will include direct microbial enumerations and phospholipid fatty acid (PFLA) analyses. Sequencing clone libraries from DNA extracted from the samples and performing phylogenetic analyses on these sequences will characterize the microbial populations. Exoenzyme assays *in situ* will also be performed to measure metabolic activity. Finally, analytical XRF and X-ray diffraction (XRD) will be performed to further characterize the concentrations and phases in which biologically important elements are present.

Acknowledgements. This work was carried out at the Jet Propulsion Laboratory, California Institute of Technology, under a contract with the National Aeronautics and Space Administration and at the NASA Langley Research Center.

REMOTE SPECTRAL IMAGING OF GEOLOGIC FORMATIONS AND ZOETIC RESIDUES WITH ACTIVE EXCITATION DIODE ARRAYS. Robert A. Lodder[1*] and Gilbert V. Levin[2] . A123 Advanced Science and Technology Center, University of Kentucky, Lexington, KY 40506-0286. 2. Spherix Inc., 12051 Indian Creek Court, Beltsville, MD 20705

The 2009 Mars Science Laboratory Mission will use a rover to seek zoetic evidence ranging from fossils of extinct organisms to current habitats that could support life. However, Martian conditions impose severe limits on the number of sites that may be visited. The authors have proposed a remote-sensing experiment, SEARCH (Scan for Extinct Astrobiological Residues and Current Habitats) to be placed on the rover to find promising sites to visit for its detailed analyses, while SEARCH obtains it own significant scientific data (see Fig. 1). Small and low-powered, SEARCH senses evidence of extinct life, water and other targets of interest at distances of 1-10 m or greater. This is accomplished by a system of spectral imaging originally designed to search for remnants of biofilms in pharmaceutical cleaning validation. An array of UV/visible/ near-IR laser light frequencies aimed at a target reflects unique profile signatures for matching against a data bank of potential target specimens (see Fig. 2). Evidence sought would include residual forms of life, such as fossils (including extinct biofilms), water, amino acids, carbohydrates and other organic matter. SEARCH could be operated at night when conditions are ideal for spectral imaging.

While SEARCH would be developed to meet the dual Mars 2009 objectives of looking for evidence of extinct life and current habitats, the method can be enhanced to look for extant life in future missions. Phototrophic endolithic biofilms, such as cyanobacteria, live on rocks where water is available only intermittently. These cells produce mycosporine-like amino acids (MAAs) along with other compounds to serve as UV screens. Detection of these molecules can serve to identify biofilms. Initial work on this enhancement of SEARCH has begun. Figure 3 shows the detection of a living biofilm, and tracks its growth over time.

Fig. 1. Proof of concept robotic model of SEARCH instrument

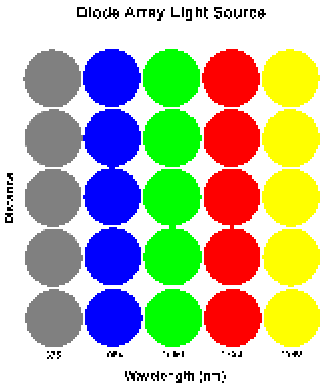

Fig. 2. SEARCH diode array light source

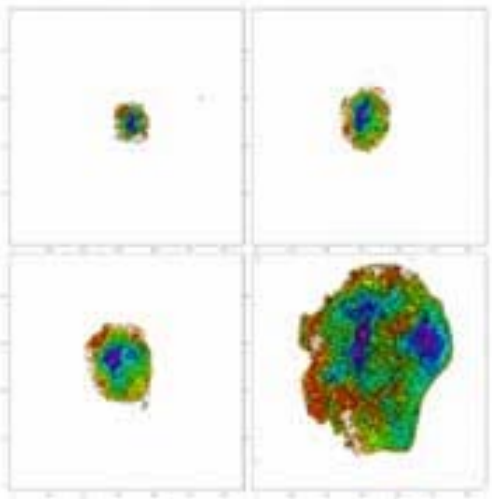

Fig. 3. BEST Contour plots of *Gloeocapsa* growing on 15 cm x 15 cm limestone wafer as seen at 9, 19, 37 and 56 days.

ENZYME-CASCADE ANALYSIS OF THE RIO TINTO SUBSURFACE ENVIRONMENT: A BIOSENSOR DEVELOPMENT EXPERIMENT.

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The EndoSafe Portable Test System (PTS), designed & developed by Charles Rivers Laboratories, Inc. (Charleston, SC) is a portable instrument that was designed to perform analysis of enzymatic assays related to rapid assessment of microbial contamination. The enzymatic cascade of Limulus Amebocyte Lysate (LAL) is known to be one of the most sensitive techniques available for detection of gram negative bacteria, yeasts and molds (future developments will include gram positive bacteria and archea), enabling the PTS to be evaluated as a potential life detection instrument for *in situ* Astrobiology missions. In the fall of 2003 subsurface rock samples from the Mars Astrobiology Research and Technology Experiment (MARTE) ground truth drilling campaign were analyzed using the LAL enzyme assay to determine if the PTS would be able to detect indigenous bacteria in the Rio Tinto subsurface. The Rio Tinto River system is an extreme environment that maintains acidic conditions and high iron and other metal ion concentrations despite seasonal temperatures and fresh water influx from rainfall & tributaries. The river is located in the core of the Iberian Pyritic belt, which is one of Earth's largest massive sulfide provinces. The data collected at this site was evaluated against contamination tracers (Sodium Bromide) and other biological analysis performed during the field season. The data shows that the LAL assay detected bacterial contamination in cores that had a high level of the sodium bromide tracer and hence verified that those cores were compromised by surface contamination due to the drilling process. The LAL assay also detected microorganisms in cores that had little to no sodium bromide tracer present, suggesting that the assay was able to detect indigenous organisms. In order for this technique to be utilized for *in situ* Astrobiology missions it will be integrated with an autonomous sample extraction and preparation system called the Biological Sample Extraction and Detection System (BSEDS). In 2005 the culmination of the MARTE project will be a high fidelity Mars drilling simulation and the BSEDS instrument will be one of the technologies included in the analysis suite.

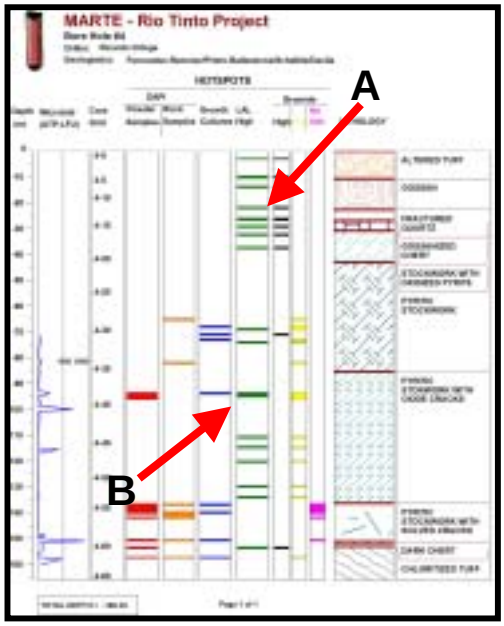

Figure 1. LAL analysis correlates well with the Bromide spiked contamination control tests (A) as well as DAPI staining and growth in culture (B).

MULTI-LAYER MICROFLUIDIC DEVICES FOR AMINO ACID BIOMARKER ANALYSIS: THE MARS ORGANIC ANALYZER

Alison M. Skelley, James R. Scherer, Andrew Aubrey[1], Pascale Ehrenfreund[2], Jeffrey L. Bada[1], Frank J. Grunthaner[3] and Richard A. Mathies

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Sensitive amino acid composition and chirality analysis has been achieved using the Mars Organic Analyzer (MOA), a portable microfabricated capillary electrophoresis (CE) instrument [1]. The microdevice consists of a four-layer sandwich structure combining glass CE separation channels, microfabricated pneumatic membrane valves and pumps, and a nanoliter fluidic network [2]. The portable MOA instrument integrates all high voltage CE power supplies, pneumatic controls, and fluorescence detection optics needed for field operation. Depending on the injection method, the concentration sensitivities range from μM to 0.1 nM for amino acids, corresponding to part-per-trillion sensitivity [3]. The MOA has been used to analyze soil extracts from the Atacama Desert; an increasing level of amino acids up to 50 ppb is detected in the north-south transect corresponding to increased precipitation levels. Field tests in the Panoche Valley CA successfully detected amino acids at the 5-to-100 ppb levels in jarosite, a key sulfate-rich mineral associated with liquid water recently detected on Mars. These results demonstrate the use of the MOA to perform high sensitivity in situ amino acid biomarker analyses in the field on samples relevant for Mars exploration. See http://astrobiology.berkeley.edu.

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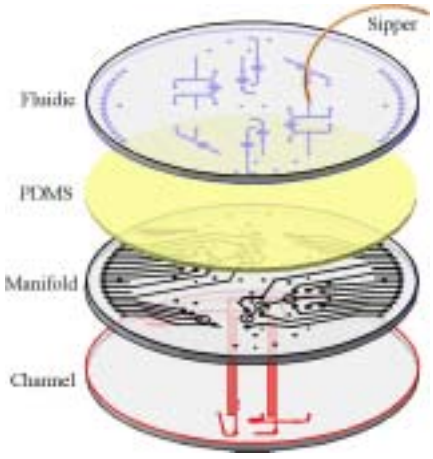

Figure 1. Microfabricated wafer for sample preparation and amino acid analysis. The 100-mm diameter microfabricated wafer stack is composed of a 4-layer sandwich of glass and PDMS to create channels and pumping structures.

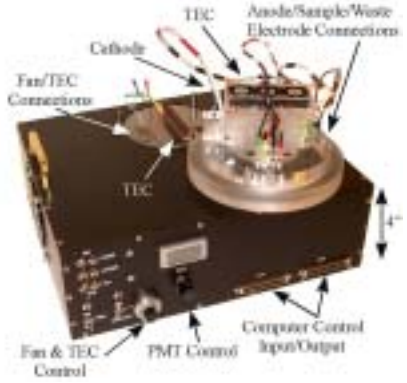

Figure 2. The Mars Organic Analyzer (MOA). The portable CE instrument, measuring 4" x 10" x 12", integrates all necessary pneumatic actuation, high voltage power supplies and confocal optics for laser excitation and fluorescence detection.

LAB ON A CHIP APPLICATION DEVELOPMENT FOR EXPLORATION

Lisa Monaco[1], Scott Spearing[1], Andy Jenkins[1], Wes Symonds[1], Derek Mayer[2], Edd Gouldie[2], Norm Wainwright[3], Marc Fries[4], Jake Maule[4], Jan Toporski[4], and Andrew Steele[4]

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NASA's Marshall Space Flight Center (MSFC) Lab on a Chip Application Development (LOCAD) team has worked with microfluidic technology for the past few years in an effort to support NASA's Mission. In that time, such microfluidic based Lab-on-a-Chip (LOC) systems have become common technology in clinical and diagnostic laboratories. The approach is most attractive due to its highly miniaturized platform and ability to perform reagent handling (i.e., dilution, mixing, separation) and diagnostics for multiple reactions in an integrated fashion. LOCAD, along with Caliper Life Sciences has successfully developed the first LOC device for macromolecular crystallization using a workstation acquired specifically for designing custom chips, the Caliper 42. LOCAD uses this, along with a novel MSFC-designed and built workstation for microfluidic development. The team has a cadre of LOC devices that can be used to perform initial feasibility testing to determine the efficacy of the LOC approach for a specific application. Once applicability has been established, the LOCAD team, along with the Army's Aviation and Missile Command microfabrication facility, can then begin to custom design and fabricate a device per the user's specifications. This presentation will highlight the LOCAD team's proven and unique expertise that has been utilized to provide end to end capabilities associated with applying microfluidics for applications that include robotic life detection instrumentation, crew health monitoring and microbial and environmental monitoring for human Exploration.

SAMPLE PREPARATION SYSTEMS DEVELOPMENT FOR THE MICROARRAY ASSAY FOR SOLAR SYSTEM EXPLORATION (MASSE) PROJECT. Molly Potter[(1)], Kristin Showalter[(1)] and Marc D. Fries[(2)]*, [(1)]Embry-Riddle Aeronautical University, Daytona Beach, FL 32114, [(2)]Geophysical Laboratory, Carnegie Institution of Washington, Washington D.C. 20015-1305, *Contact author email: m.fries@gl.ciw.edu

Abstract

Life detection missions present special instrumentation challenges since they must detect minute quantities of specific molecules under hostile environmental conditions with minimal uncertainty. The Microarray Assay for Solar System Exploration (MASSE) project will utilize antibody microarray techniques to interrogate regolith and rock samples for a broad suite of biomarker molecules using wet-chemistry techniques already widely used on Earth. This technique is applicable to sample materials from a wide range of extraterrestrial environments but requires liquid extraction methods to liberate biomarkers from matrix material for analysis. The MASSE sample handling system must start with a solid rock or regolith sample and prepare a concentrated, aqueous extractant solution for analysis. Considerable uncertainty exists in the effects of salinity, pH, temperature, and other parameters upon the extraction and concentration procedures. A course of experimentation is underway to characterize sample handling efficiency as well as precisely understand the effects of a wide range of environmental variables. Sample handling is critical to mission success and must be thoroughly understood in a laboratory setting even before hardware design can commence. An overview of the equipment, systems, and experimental procedures necessary for the development of a sample handling system will be presented and discussed.

The Inchworm Deep Drilling System For In Situ Investigations of Martian Subsurface Aquiferous Zones. T.Myrick[12], S.Frader-Thompson1[13], J. Wilson[14], S. Gorevan[15], [1]Honeybee Robotics, 204 Elizabeth Street, New York, NY 10012, [2]Myrick@HoneybeeRobotics.com, [3]Frader-Thompson@HoneybeeRobotics.com, [4]Wilson@HoneybeeRobotics.com, [5]Gorevan@HoneybeeRobotics.com

In reviewing the Astrobiology Roadmap [1] and its objectives, as well as the goals and investigations sanctioned by the Mars Exploration Program Analysis Group (MEPAG)[2], an explicit need emerges for a vehicle to bring instrumentation to the subsurface water ice deposits and horizontal aquiferous zones of Mars. Recent findings indicating that water in the liquid phase on Mars may exist at 100 meters or more below the surface points out the need to obtain a means of practical deep subsurface access. The Inchworm Deep Drilling System (IDDS) addresses that explicit need. The IDDS is a low mass, novel subsurface access technology that is currently being developed with the support of NASA HQ. The IDDS is endowed with the potential to drill tens to hundreds of meters if tethered to a base platform on the surface, or if un-tethered, could access locations several kilometers below the surface of Mars. Yet the IDDS does not require the onerous mass and power of terrestrial drilling systems. An un-tethered, self-contained IDDS may utilize Radioisotope Thermoelectric Generators (RTGs) for fully autonomous operation at depths below a few to several hundred meters. By keeping one set of borehole wall shoes firmly secured to the borehole wall, on either the forward or aft section, the other section is able to expand or retract (like an inchworm), allowing the IDDS to drill into competent rock or ice and traverse the borehole. This method of locomotion is independent of gravity and allows the IDDS, tethered or un-tethered, to traverse the borehole to the surface for cuttings removal, external sample analysis, or to accommodate sample return. The IDDS is capable of delivering an instrument suite to candidate subsurface targets to perform in situ analyses of biogenic mineralogy and possible liquid water zones in and around assumed aquiferous seepage channels, paleoenvironments [3], and water ice deposits at moderate depths of 100 meters or more. The instrument suite could include the combination of mission specific borehole analytical instruments, such as microscopic and multi-spectral cameras and various spectrometers, with innovative stratigraphy-maintained sampling and sample manipulation mechanisms, such as a Mini-Corer-like coring bit and a method of delivering cuttings and cores to the onboard instrumentation or storing the samples for delivery to the surface. Honeybee Robotics believes the drilling mechanisms and subsystems, including the instrument suite, could be integrated into a compact and robust system (on the order of two meters long and twelve centimeters in diameter, however, the size depends heavily on the number and type of instruments and the sample acquisition and delivery methods required). The proposed technology maturation is funded under NASA's Planetary Instrument Definition and Development (PIDD) and Astrobiology Science and Technology for Exploring Planets (ASTEP) programs.

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Thermal properties of saline environments: potential targets for detecting signatures of past or present life on Mars.

Author: Stanley Scher, P.O. Box 9456 Berkeley, CA 94709

Abstract: The search for water on Mars is central to our understanding of pre-existing or extant life on that planet. Several lines of evidence suggest that substantial quantities of water may have existed, or currently exist as ice on the surface or subsurface of Mars.

On Earth, approximately half of all inland waters are saline. By analogy, we can assume

that at least some of the water on Mars may be associated with sites containing salts. Concentrated salt solutions—brines—form by evaporation, or interaction of salt deposits with water-ice. High salinity brines not only depress the freezing point of water; they also serve to stabilize the heat capacity.

The heat capacity of ice is less than half that of liquid water. Accordingly, we can expect saline brines or other salt-rich aqueous environments to remain liquid over a wider range of temperatures and to exhibit greater thermal stability compared with analogous regions lacking or depauperate in dissolved salts.

We propose here that the lower freezing point and high heat capacity of saline waters may provide a more stable medium for salt-tolerant microorganisms on Mars or elsewhere. This combination of thermal properties may represent a decisive factor in the selection of target sites for detecting evidence of past or present life on Mars, or by analogy, sub-freezing polar environments on Earth.

Submitted to the MARS ASTROBIOLOGY SCIENCE AND TECHNOLOGY WORKSHOP, 8-10 SEPTEMBER, CARNEGIE INSTITUTION OF WASHINGTON

IN-SITU MEASUREMENTS OF IONIC MOTION DIRECTLY IN PLANETARY SOILS

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The search for extant life is one of NASA's most important goals. The existence of life, even in the most inhospitable terrestrial environments suggests that biosignatures of life may be ubiquitous provided we know what to look for.[1] Empirical evidence suggests that life, as we know it, requires an adequate supply of water. Recent Microscopic Imager and mineralogical data from Mars Opportunity and Spirit indicates a watery past. Mars Odyssey neutron and γ-ray orbital data are also being interpreted as indicative of the continued presence of near-surface water at mid-latitudes, albeit in relatively small amounts compared to most terrestrial environments.[2] Measuring ionic motion of these water-bearing soils will provide important information on the state of the soil and its capacity as a medium for harboring life. Moreover, terrestrial studies have demonstrated a close relationship between the depositional environment and the physical and chemical properties of the soils.[3,4] Such measurements would, therefore, also be important in determining the importance of chemical weathering and cementation phenomena.

Analytical laboratories in development for planetary exploration while, in principle, capable of such measurements, disturb the soils in gathering and processing the samples, thereby altering the chemical state of the native soil. The required consumables of these laboratories further limit the number of such measurements that could be made, handicapping their ability to perform large area surveys. We report here some results on the viability of an analytical instrument that eliminates the "laboratory" by performing measurements directly in the native planetary soils (see Figure 1). The data indicate that an appropriately configured instrument is capable of measuring ionic motion under the < 3 % water reserve identified by GRS. While further studies are needed to confirm the robustness of this approach under other environmental conditions, these initial measurements are extremely encouraging on its viability.

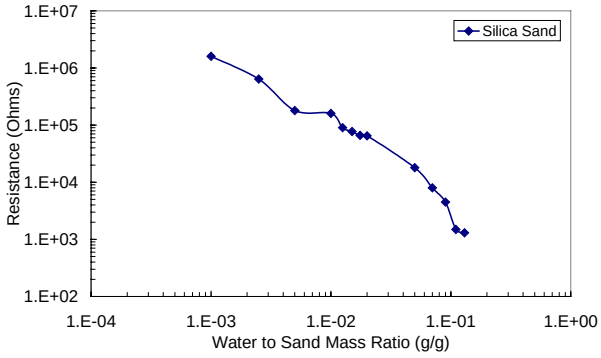

Figure 1: Electrical conductivity of fine silica sand mixed with varying amounts of water containing 100mM KCl solution.

Acknowledgements: The work described in this paper was performed under a contract with the NASA, Astrobiology Science and Technology Instrument Development Program managed by Dr. Michael Meyer. We are also indebted to G. Kuhlman, of JPL, T. Sant and E. Brizendine of Univ. Nevada at Reno and Dr. M.Schaap of the USDA Salinity Laboratory and UC Riverside for their help in the experiments.

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SMILE (Specific Molecular Identification of Life Experiment): A Family of Molecular Recognition Sensor Instruments for Robust Detection of Life in the Solar System.

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The detection of life elsewhere in the Solar System is a central component of the astrobiology discipline. One approach to identify life is via (i) detection of molecular evidence of past or present life in the form of organic molecular biomarkers together with (ii) an understanding of the contextual abiotic organic environment in which biomarkers may be found. The latter is also of interest in the exploration of other bodies such as comets and asteroids.

For environmental and medical applications on Earth that desire *in situ* detection and characterisation of trace organic markers, molecular receptor based sensor technologies are receiving much attention. The current trends of multi-analyte sensor arrays, μ-fluidic and μ-system approaches, and transducer readout technology development is making remote analytical systems for astrobiology and planetary exploration a realistic practicality.

For life detection applications and information-critical terrestrial applications such as bio / chemical treat detection, the requirement for analytical robustness, *i.e.* lack of false positives / negatives, is vital. At present, the academic / commercial examples of molecular receptor based devices do not offer the level of analytical robustness desired for current life detection scenarios.

We have proposed an approach to molecular recognition sensor arrays, *i.e.* biosensors arrays, for the *in situ* detection of trace organic biomarkers that addresses the practical issues of analytical and physical robustness of such approaches. Thus, the "*Specific Molecular Identification of Life Experiment*" (SMILE) is based upon the concept of the integration of diverse molecular sensor materials in array formats and with multiple transduction readout technologies. Such an approach gives an information rich output both in the context of immediate sensor operation and contextual sample information. Such information will allow both analytical determination of the presence of given biomarkers, or classes of biomarkers, and a confidence level in the measurements to identify false readings.

The present SMILE concept has three major embodiments comprising:

- "*lite*" version (~ 500 g) involving multiple single-use μ-fluidic capillaries containing immobilised arrays of molecular materials appropriate for molecular recognition and other sensing assays with real-time fluorescent readout via optical evanescent excitation
- "*pro*" version (~1,500 g) involving multiple-use array of molecular sensing materials integrated on the surface of a hybrid optical and electrochemical readout transducer with surface plasmon resonance detection of interfacial refractive index and evanescent fluorescence excitation of molecular recognition assays together with electrode arrays for electrochemical measurement of additional sample parameters
- "*pro plus*" version (~ 3,000 g) that includes capillary electrophoresis and PCR modules

Present studies include integration of molecular sensing materials, mission and operational stability studies for molecular recognition materials, assessment of antibody and molecular imprinted polymer molecular recognition materials.

A comprehensive plan for drilling in Martian permafrost in search of life.

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If life ever existed on Mars, a key question is the genetic relationship of that life to life on Earth. To determine if Martian life represents a separate, second genesis of life requires the analysis of organisms, not fossils. Ancient permafrost on Mars represents one potential source of preserved, albeit dead, Martian organisms. Strong crustal magnetism in the ancient heavily cratered southern highlands between 60 – 80^{o}S and 180^{o}W indicate what may be the oldest, best preserved permafrost on Mars. Drilling to depths of 1000 m would reach samples unaffected by possible warming due to cyclic changes in Mars' obliquity. When drilling into the permafrost to retrieve preserved Martian organisms, it is necessary to take special precautions to avoid the possibility of contamination. On Earth, and possibly on Mars, it is impractical to sterilize the entire drill. Even if initial sterilization is possible, contamination of deep layers with surface material must be considered. It is clear that the exterior of the samples taken from aseptic drilling methods are contaminated by the drilling environment. However because of the impenetrability of frozen cores, it is likely that the inner core of the sample is sterile and uncontaminated by the drilling environment. Problems with current drilling methods, techniques to trace the cleanliness of samples collected, and complications with maintaining an aseptic environment are considered here in the context of Martian permafrost drilling. . Suggestions for future research on drilling methods and tracer technique developments are considered. Mars analogs on Earth, such as the Canadian Artic and Siberia, provide an suitable environment for testing such technologies. Advances made on Earth in aseptic drilling form the basis for the design of a Mars permafrost drill.

ACTIVITY OF THE ENZYME HYDROGENASE AS A PROXY FOR HYDROGEN METABOLISM IN DEEPLY BURIED SEDIMENTS, AND AS A DIAGNOSTIC TEST FOR LIFE.

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The microbial communities that inhabit deeply buried sediments are of considerable interest to geochemists and astrobiologists because they are important in terrestrial biogeochemical cycles, and can serve as models for extraterretrial ecosystems. The metabolic rate of these communities can be extremely low and is therefore difficult to measure directly; this issue is a current challenge in terrestrial deep biosphere studies, and is relevant to life-searching space missions.

Hydrogen gas (H_2) is a key intermediate in anaerobic metabolism, because it is produced and utilized by a large number of organisms, and because it acts as a metabolic link between microbes with very diverse substrate requirements. The importance of H_2 suggests the hypothesis that enzymatic (catalytic) activities related to its metabolism might be good indeces of overall microbial community metabolism. Thus, an assay to measure H_2-related enzymatic activities would be very valuable for deep biosphere studies, and if sensitive enough, it might be useful as an exploratory tool for the presence of life. To test this hypothesis, we are developing a radiotracer assay for *Hydrogenase*, an enzyme that all H_2-producing and H_2-consuming microbes possess.

The method relies on the known ability of hydrogenase to catalyze an isotopic exchange between dissolved H_2 and the hydrogen atoms in water. This property has been exploited for water column measurement of hydrogenase with tritium gas as tracer (Schink et al, 1983), but has never been applied to sediments. To simulate sediment with low microbial activity, a dilute slurry of local coastal sediment is used as the test material. The slurry is incubated under a tritiated H_2 headspace, sampled over time, and scintillation counted. With high partial pressure of H_2 (0.5 atm) the rate of accumulation of tritium in the sediment slurry is proportional to the amount of enzyme present. Currently, the assay can measure hydrogenase activity routinely in a 1:10,000 sediment dilution. Work in progress is focused on further increasing the sensitivity of the assay. The set-up has been adapted for future deployment in an International Ocean Drillig Program expedition. With appropriate modifications (e.g. use of deuterium mass spectrometry), this simple and sensitive technique might be applicable to planetary exploration.

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Modular Assays for Solar System Exploration

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With the advent of a new era of Astrobiology missions to search our solar system for evidence of life we would like to update the community on the use of protein microarrays for detection of viable and fossil life biomarkers on Mars. We successfully proposed this instrument in 1999 to NASA and ESA for the now defunct 2005 mission (under the title MILDI) and have recently received NASA ASTID funding. Science and engineering definition have been ongoing since 1999, culminating in system designs and breadboards that are now being tested under ASTID funding.

It is imperative that if biotechnology techniques such as protein microarrays or other similar types of instruments be used for life detection on Mars that proof of concept science to show the compatibility of the probes (i.e. antibodies), fluorophores, reagents, reactions and microfluidic chips for space flight and Martian environments. In summary over the last two years the MASSE project has:

- Developed an initial custom protein array for the detection of viable life marker molecules. This chip has been tested using several extraction methods and using spiked Martian regolith.
- Developed a fossil antigen marker chip that has successfully detected antigenic activity in samples up to 65 million years old.
- Developed and tested antihopane antibodies for fossil life detection. This work is ongoing.
- Verified the antibody binding reaction kinetics in Martian and 0 G testing.
- Tested 25 separate extraction protocols for use in a microarray microfluidic device.
- Demonstrated gram negative, positive and archaeal cell lysis in microfluidic channels.
- Built and are currently testing a prototype extraction device for use with simulated Martian regolith.
- Demonstrated the use of the Limulus Amoebocyte Lyase test as a verification test for bacterial detection by protein microarrays and indeed shown how the enzymes involved may be used in a microarray format.
- Designed microfluidic systems for all the steps to prepare a sample for inoculation onto a microarray.
- Built a breadboard optical detection system and a design for a miniaturized unit.
- Currently undertaking radiation exposure experiments for fluorophores and microarrays and have tested the longevity of microarrays for use in a Mars instrument.

We will present a cross section of these results and show how we have field tested the protocols for microarray analysis and show how the science definition studies outlined above will aid in instrument definition and development for the ESA Exomars opportunity.

In-Field Testing of Life Detection Instruments and Protocols in a Mars Analogue Arctic Environment

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During August of 2003 an expedition including 19 scientists tested various equipment at a series of hotsprings sites on the island of Svalbard. This island is considered a Mars analogue environment due to the presence of hot springs, carbonate terraces and volcanic activity which have produced carbonate rosettes similar to those found in ALH84001. The goal was to test 4 portable instruments for their robustness as field instruments for life detection (for future human missions to Mars), to assess the Mars analogue environments for signs of life, to refine protocols for contamination reduction and to understand the effects of transport on sample integrity by assessing bioloads immediately in the field and then comparing these with laboratory measurements made after transportation.

The instruments used in this investigation were; ATP luminometry which assays for ATP as a measure of metabolic activity; Scalar DG2 hand held digital microscope which enables digital microscopy images to be captured at magnifications of up to x200. Charles River Endosafe unit which uses the Limulus Aemebocyte Lysate assay (LAL) and Mobile PCR, which is manufactured by MJ research.

We were able to detect and quantify bacterial load from several sites including endolithic communities from travertine dry terraces at the site. We have been able to use functional gene analysis in the field to rapidly constrain the types of bacterial activity being undertaken within the hotsprings site. We refined one suite of techniques and protocols to enable primary life detection and characterisation in the field. These techniques could also be used in support of trials of flight hardware being developed to detect microbial life on Mars or elsewhere.

Ackowledgements
The entire AMASE team and crew of M/S Polarsyssel especially Tori Hoehler who spotted the endoliths. NAI Wes Huntress, Mike Meyer and the ASTEP program.

Subsurface Sampling and Sensing using Burrowing Moles

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Finding evidence for life on Mars will likely require accessing the subsurface since the Martian surface is both hostile to life and to preservation of biosignatures due to the cold dry conditions, the strong UV environment, and the presence of strong oxidants. Systems are needed to probe beneath the sun and oxidant baked surface of Mars and return samples to the surface for analysis or to bring the instrument sensing underground. Recognizing this need, the European Space Agency incorporated a small subsurface penetrometer or "Mole" onto the Beagle 2 Mars lander. Had the 2003 landing been successful, the Mole would have collected samples from 1-1.5 m depth and delivered them to an organic analysis instrument on the surface. The device called the Planetary Underground Tool (PLUTO), also measured soil mechanical and thermophysical properties. Constrained by the small mass and volume allowance of the Beagle lander, the PLUTO mole was a slender cylinder only 2 cm diameter and 28 cm long equipped with a small sampling device designed to collect samples and bring them to the surface for analysis by other instrument. The mass of the entire system including deployment mechanism and tether was 1/2 kg.

In addition to a mechanism for collecting subsurface samples, Moles can be used to carry a sensor package underground to make *in situ* measurements. The Mars Underground Mole (MUM) is a larger Mole based on the PLUTO design but incorporating light collection optics that interface to a fiber optic cable in the tether that transmits light to a combined stimulated emission Raman Spectrometer and Short Wave Infrared (SWIR) reflectance Spectrometer with sensitivity from 0.7 to 2.5 micrometers. This instrument is called the Dual Spectral Sensor and uses a Digital Array Scanning Interferometer as the sensor technology, a type of fourrier transform interferometer that uses fixed element prisms and thus is highly rugged compared to a Michaelson interferometer. Due to the size limitations of an on-Mole instrument compartment, and the availability of a tether, the sensor head, light sources, and control electronics for the instrument are on the surface. The DSS sensor is capable of sensing a wide range of minerals relevant to Mars Astrobiology objectives including hydrated minerals, clays, carbonates, sulfates, and ice. Additionally, Raman spectroscopy is effective for detecting organics. The MUM is designed to achieve a maximum depth of penetration of 5 m in Mars regolith and can be repeatedly deployed and retrieved. The ability to perform repeated sampling, combined with the low mass and power requirements, means that Moles could be incorporated into a rover mission as well as used on a stationary platform.

The Mole mechanism is a pointed slender cylinder that advances into soil by way of an internal sliding hammer mechanism. Part of the energy released by the spring-loaded hammer with each shock is transferred to the Mole casing and from there to the soil, resulting in penetration by displacing and compressing the surrounding soil. A backwards-directed impulse as a reaction to each forward shock is transferred via a suppressor mass against a second weaker spring allowing forward motion without requiring reactive forces provided by the lander. The Mole tip can be opened to collect soil samples. The Mole casing is tethered to a supporting mechanism that supplies power. Components supporting the Mole on the surface include a launch tube, tether reel and winch for pulling in tether, in addition to the tether itself.

Mars Analog Rio Tinto Experiment(MARTE): An experimental demonstration of key technologies for searching for life on Mars

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The discovery of near surface ground ice by the Mars Odyssey mission and the abundant evidence for recent Gulley features observed by the Mars Global Surveyor mission support longstanding theoretical arguments for subsurface liquid water on Mars. Thus, implementing the Mars program goal to search for life points to drilling on Mars to reach liquid water, collecting samples and analyzing them with instrumentation to detect *in situ* organisms and biomarker compounds. Searching for life in the subsurface of Mars will require drilling, sample extraction and handling, and new technologies to find and identify biomarker compounds and search for living organisms.

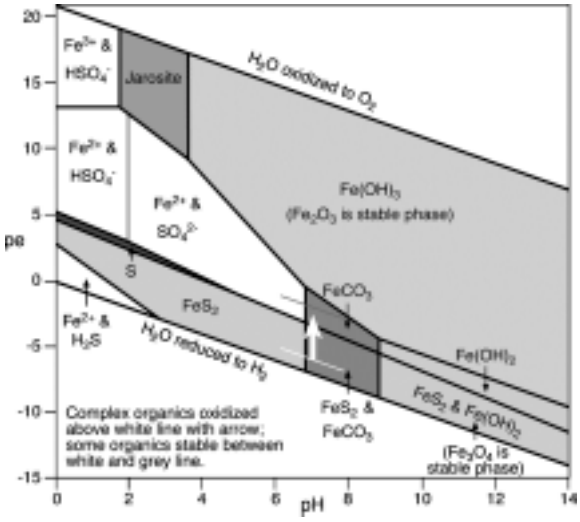
MARTE (Mars Analog Rio Tinto Experiment) is a field experiment to search for evidence of a subsurface biosphere analogous to one that could be found on Mars. MARTE is developing drilling, sample handling, and instrument technologies relevant to searching for a Martian biosphere, and demonstrating them in a field test at a site with a Mars-analog subsurface biosphere. The mission simulation will employ a drilling system developed by Honeybee robotics for future use on Mars that produces 25 cm core segments at 2.5 cm diameter while operating on low power without the use of drilling fluids. An automated Core and Sample Handling facility will extract cores from the drill and pass them to a suite of instruments on the surface. Cores are examined by remote sensing instruments including a panoramic context imager, microscopic imager, and a visible-near infrared hyperspectral imager. A sterile swab from each core is examined using ATP luminometry for a quick-look indication of the presence of living organisms. Logging instruments deployed in the borehole include a camera, magnetic susceptibility meter, and raman spectrometer. A science team located at remote operations centers analyzes the data from the logging instruments and selects core locations to sample with a life detection instrument located onboard the lander. Once subsample locations are chosen, a subsection of core is cut out, crushed, and then placed into the SOLID prototype life detection instrument for further processing. The SOLID is a portable automated instrument that uses DNA and protein microarray technology to detect microorganisms as well as their metabolic products. The instrument is capable of sensing many kinds of biochemical compounds (nucleic acids, proteins, polysaccharides, etc) using microarrays printed with DNA, antibodies or any other protein or molecule able to recognize and bind specifically to them. A Mars drilling mission simulation will be performed in June 2005 that includes interpretation of drill mission results by a remote science team in a blind test. This simulated drill mission is augmented by additional "ground truth" utilizing more conventional methods for drilling, sample handling, and laboratory analysis to explore for a subsurface biosphere at the field site. The MARTE project achieves exploration of an uncharacterized underground ecosystem of key relevance to Astrobiology and the search for life on Mars, while also developing and demonstrating technology needed in the next phase of Mars exploration.

Acknowlegements: MARTE is funded by a grant from the NASA ASTEP program.

POOR PRESERVATION POTENTIAL OF ORGANICS IN MERIDIANI PLANUM UNIT P2 SEDIMENTARY ROCKS. Dawn Y. Sumner, Geology Department, University of California, Davis, CA 95616, sumner@geology.ucdavis.edu.

Issue. New data from MER rover Opportunity, which landed in Meridiani Planum P2 sedimentary rocks as mapped by Hynek et al. [2002], demonstrates substantial water flow and water-rock interactions in pre-3 Ga sediments containing substantial sulfate evaporites, jarosite, and hematite. These data have increased excitement over the exploration for an early Martian biosphere. However, this search needs to be carefully directed by the preservation potential of possible biosignatures, including organic compounds. The abundance of Fe(III) in unit P2 indicate that organic compounds have a poor preservation potential in these sedimentary rocks.

Fe-S-C Chemistry and Rocks. Organic compounds consist of variably reduced carbon, whereas hematite and sulfate minerals contain oxidized iron and sulfur. These 3 phases are not in thermodynamic equilibrium (figure) and are not expected to be present in the same rocks. The kinetics of sulfate reduction to sulfide are slow, however (e.g. billions of years at pH~>4; Ohmoto and Lasaga, 1982), so the preservation of organics in sulfate-rich sediments is kinetically reasonable. In contrast, Fe(III) reduction to Fe(II) is rapid and strongly coupled to oxidation of organics in water and sediments [e.g. Lovley et al., 1981], in part through the catalysis of oxygen-derived free radicals [e.g. Stumm and Morgan, 1996]. This reaction potential is commonly observed in rocks. Hematite-rich facies lack organics whereas siderite (Fe(II) carbonate) facies are commonly associated with preserved organic compounds [e.g. Kaufman et al., 1990]. Where organics are preserved in acid lake deposits, hematite has been reduced to soluble Fe(II) in "reduction spots" [e.g., Benison and Goldstein, 2001]. Organic inclusions within associated sulfates have not been reported. Similarly, iron-rich concretions only contain organic carbon when formed of pyrite (Fe(II) sulfide) or siderite; hematite concretions form around oxidized zones rather than reduced zones [e.g. Chan et al., 2000].



Implications for P2. Hematite concretions are excellent for indicating extended water-rock interaction, but complex organic compounds that would act as biomarkers are not likely to be preserved in sediments rich in Fe(III), even if they were abundant when the sediments were deposited. Identification of reduced iron minerals could indicate redox reactions consistent with organic decay (but not requiring organic decay). However, in the absence of strong evidence for specific conditions that would isolate organics and Fe(III) from each other in these rocks, *in situ* characterization of organic compounds in P2 rocks is unlikely to produce positive results.

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Feasibility of Enzymatic Assay for ATP as an Indicator of Subterranean Microbial Life on Mars.

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With the stunning success of the recent Mars Rover missions, interest in the detection of life on the Red Planet has been renewed. Specifically, evidence in the geology of the Martian surface seems to indicate that water was once abundantly available. It has been postulated that although the surface of Mars is currently cold and barren, below the surface of the planet lie subterranean water sources. It is this water table that presents the most likely place for microbial life on Mars. This poster will discuss a method to reach this underground water, and will focus on the use of an enzymatic assay for the detection of the biochemical marker adenosine triphosphate (ATP).

In the search for living organisms on other planets, it is useful to consider common traits of life as we know it. The ubiquitous energy molecule ATP provides an excellent target and indicator of cellular life forms. A sensitive assay exists for ATP that takes advantage of a natural enzyme, firefly luciferase. This enzyme, when combined with the substrate luciferin and two other cofactors, catalyzes a reaction that efficiently releases light (quantum efficiency >90%). The use of this enzyme represents a simple, selective, and sensitive means to determine the presence of ATP, and in turn, living organisms. However, the implementation of this assay on or below the surface of Mars is not as straightforward as performing the assay in a laboratory. As was stated previously, any microorganisms are likely to be concentrated in subterranean reservoirs of liquid or frozen water. Thus, to effectively test for these microorganisms with the luciferase assay, one must first drill through frozen layers. Also, since this water has slowly receded through rock layers, it is likely to be highly saline, with high contents of sodium, calcium, and other inorganic ions. This brine solution presents a challenge in that the enzymatic nature of the ATP assay can be adversely affected by the salinity of the surrounding liquid. Steps must then be taken to mitigate the effects of the brine on the assay.

The focus of this poster will be on the development of a method to employ the firefly luciferase ATP assay for the detection of microorganisms on Mars. Strategies to overcome interference effects of the concentrated brine solution will be discussed, along with preliminary results in this area. Our strategies include two main methods, the first of which involves removing bacteria from the brine solution, followed by bacterial lysis in a buffer compatible with the luciferase enzyme. The second strategy attempts to simplify the analysis by working directly in the brine medium. To protect the enzyme, micellar surfactants and other chemical enhancers can be added in order to shield the enzyme from the negative effects of the brine components. An overview of these strategies along with conceptual drawings of how the equipment can be integrated into a compact module will be provided.

MAGNETOELECTRONIC MICROARRAY DETECTION OF MAGNETICALLY LABELED BIOMOLECULES. Cy Tamanaha, Christina Cole, Shawn Mulvaney, Jack Rife and Lloyd Whitman, Naval Research Laboratory, Washington, DC 20375, email cy.tamanaha@nrl.navy.mil.

Abstract. Many of the current microarray technologies, particularly DNA arrays, rely on optical detection methods such as chemiluminescence, fluorescence, or colorimetric assays. We have developed a fundamentally different technique to quantitatively detect and identify biological molecules whereby captured molecules (e.g. DNA, proteins) are labeled with paramagnetic microbeads and subsequently detected by an array of giant magnetoresistance (GMR) magnetic field sensors embedded in the substrate (the Bead ARray Counter, or BARC chip).[1] A complete analytical system called the compact Bead Array Sensor System (*cBASS*), which integrates the electronic instrumentation with a compact fluidics system,[2] is being developed around this technology. Although our technology has its roots in biological warfare agent detection, it has many other potential applications, including biomedical research, point-of-care diagnosis, high-throughput drug screening, and environmental monitoring.

In its basic configuration, our biosensor system uniquely combines a receptor microarray, paramagnetic microbeads, GMR magnetic field sensors, and microfluidics to detect and identify biological molecules (see Figure 1). At its core is a BARC microchip containing the GMR sensor array. The assay is performed on the microchip in a flow cell using a hybrid macro-microfluidics system. Distinct receptor probes are immobilized above each sensor. Complementary target molecules (ligands) in a sample are captured by highly-specific ligand-receptor interaction on the chip, and are then labeled with paramagnetic microbeads. Controlled microfluidic forces are then applied to remove non-specifically bound microbead labels over the sensor area. Finally, the remaining magnetic labels are detected by the GMR sensors, providing a quantitative measure of the target concentration. The challenges of achieving effective assay and instrumental sensitivities in addition to the interdisciplinary effort required in making a complete, integrated sensor system based on this technology will be discussed.

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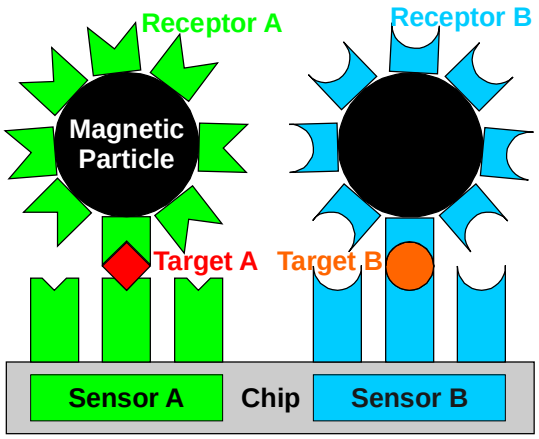

Figure 1. Generic illustration of magnetic labeling of target ligands captured onto a solid substrate in a sandwich configuration using specific biomolecular ligand-receptor recognition.

Searching for life on Mars and beyond: Using Fluorescence Biological Analysis (FBA) to Examine the Subsurface of Europa for Signs of Life.

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Resent evidence suggest that water was once present in large amounts on Mars. In fact, Mars may have been habitable. Another location that is high on NASA's search for life outside of our earthly parameters is Europa, one of Jupiter's Icy Moons. Two important components essential to life include the present of water and a source of energy. As a result of these two factors, multiple theories suggest the possibility of life on Europa. The surface of Europa is composed of thick layers of ice produced by the combination of an underwater ocean and extremely low temperatures. The underwater ocean is a result of expected volcanic activity along the ocean's floor. The area between the thick layers of ice and the underwater ocean is potential breeding ground for a host of bacteria, cells, and biomolecules. The fluorescence biological analysis system is a direct result of cutting edge technology and innovative biological research. This system consists of an orbiter and a penetrator with various tools and instruments needed to complete this mission. The orbiter will use jet propulsion to launch the penetrator, forcing it into the surface of Europa in order to examine the subsurface in situ. Samples of water will be collected and immediately examined using fluorescence biological analysis.
Fluorescence is a proven method used for locating organic molecules that are present within a specific region.

Acknowledgements
We wish to thank Dr. Juan Pérez Mercader for inviting La Tasha Taylor to Spain and arranging for her lodging and the Centro de Astrobiología for hosting her visit.

Microfluidic measurement of microbial bioburden by fluorescent LAL assay.

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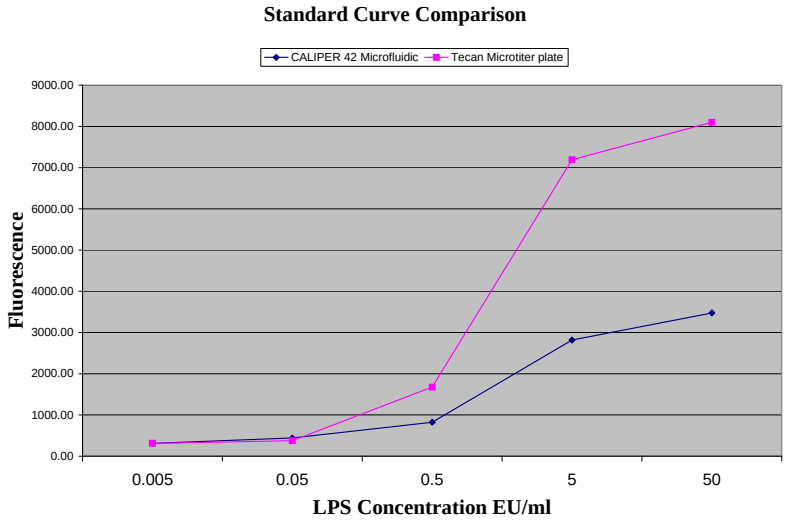
The technology to detect extremely low-level signatures of microbial life is equally important in support of Planetary Protection requirements, as well as life detection missions. Furthermore, the ability to report data near the point of collection and in near real-time is highly desirable. The use of microfluidics and other miniaturization technology will allow such portability in hand-held devices and flight instruments. While culture-based methods remain an essential tool, the 2-3 day culture period is excessively time consuming and does not address the level of non-viable organic contamination. During spacecraft assembly, rapid feedback on cleanliness levels may avoid costly delay and reliable *in situ* life detection methods will surely benefit the depth of science investigation capable of being performed on the Moon, Mars and other bodies in our Solar System.

The Limulus Amebocyte Lysate (LAL) assay is a sensitive enzyme cascade that is triggered by microbial cell wall material. Lipopolysaccharide from gram negative bacteria and beta glucan from yeast and mold can be detected as low as 10^{-13} g in less than one hour. The cascade is part of the innate immune system of the horseshoe crab, *Limulus polyphemus* in which microbes that enter its blood system through an open wound are recognized as foreign. Bacterial lipopolysaccharide (LPS), also called endotoxin, is an integral part of the outer membrane and begins the cascade by binding to, and activating the protease pro-enzyme. Techniques to discriminate whole cells from fragmented cells will be discussed. The LAL test has been adapted to use chromogenic or fluorogenic substrates we use to quantify the reaction. We report successful analysis of fluorogenic LAL reactions in a prototype microfluidic chip.

LAL reactions (Endochrome, Charles River Laboratories) were performed on a series of LPS concentration using the fluorogenic substrate Ile-Glu-Gly-Arg-AMC (Enzyme Systems) and standard endpoint analysis. Reactions were read both in a Tecan Spectrafluor Plus microplate fluorometer as well in the microfluidic chip (Caliper 42). The figure below shows the same standard LPS concentration series run in a standard Tecan microtiter plate and in the Caliper 42 microfluidic system. Despite the large difference in path length, both curves show equal sensitivity. We will continue development of the microfluidic LAL to incorporate all aspects of sample handling, dilution and analysis.



DIELECTRIC SPECTROSCOPY FOR *IN SITU* DETECTION OF MICROBIAL LIFE FORMS ON MARS. D. Warmflash[1,2], J. H. Miller, Jr.[1], D.S. McKay[2], G.E. Fox[1], and D. Nawarathna[1]; [1]University of Houston; [2]NASA Johnson Space Center, Houston, TX, email dwarmfla@ems.jsc.nasa.gov

A challenge to astrobiological investigation of Mars is to develop *in situ* instruments capable of distinguishing environmental samples or extracts containing life forms from those that do not. At the same time, the life-detection technology must not be geocentric; that is it must not be targeted to characteristics that, although specific for life, may be limited to those life forms native to Earth. Because life throughout the Cosmos, regardless of its biochemistry and/or its genetic material, must utilize a variety of charged macromolecules, we are investigating the use of dielectric spectroscopy (DS) as a life-detection tool.

In an initial study, as environmental samples, we used common soil and JSC Mars-1, a volcanic ash from Hawaii, developed for use as a Mars regolith simulant. Biologically active, JSC Mars-1 contains microorganisms and biomolecules equivalent to 10^6–10^7 cells/gram, less than common soils (which can contain quantities of up to 10^9 cells/gram). Portions of each environmental sample were left untreated, while other portions were sterilized: autoclaved for 60 minutes at 121˚ C, at 2 atm, then heated in an oven at 220˚ for 3 hours followed by exposure to ultraviolet light for 16 hours. Water extractions were then performed on sterilized and untreated soil and JSC Mars-1 samples. Extracts of untreated soil and JSC Mars-1 yielded multiple microbial strains when incubated on Luria Broth (LB) agar for 24 hours at three temperatures: 23˚ C, 30˚ C, and 37˚ C. Extracts of sterilized soil and JSC Mars-1 showed no growth at any of these temperatures, indicating that the sterilization protocol had indeed destroyed all living forms within the environmental samples.

When DS was conducted on extracts, dielectric constant and conductivity were found to be higher for sterilized samples as compared with untreated samples. We hypothesize that the sterilization protocol results in increased dielectric constant and conductivity due to lysis of cells and consequent release of charged molecules. However, the values obtained for unsterilized samples may be due not only to the presence of charged molecules but also to membrane potentials of living cells. Samples containing living cells may thus be distinguishable from those containing only macromolecules by performing DS at variable temperatures. Thus at two temperatures, 4˚ C and 37˚, we tested a suspension of the bacterium, *E. coli*, as well as three examples of large, charged biomolecules: deoxyribonucleic acid (DNA), hemoglobin (Hb), and bovine serum albumin (BSA). At 10 Hz, dielectric constant for *E. coli* suspension increased by 70% at 37˚ as compared to 4˚, while dielectric constants for DNA, Hb, and BSA increased by 28% 17%, and 49% respectively. Furthermore, DNA, Hb, and BSA used in this preliminary study were not sterile, so it is possible that the dielectric differences measured at the two temperatures for these macromolecules –although lower than the difference observed for *E. coli* – are due in part to bacterial contamination and this will be investigated.

DS may be applicable to *in situ* astrobiology studies on the surface of Mars and the variable temperature method will be tested on soil and JSC Mars-1. Additionally, other manipulations, such as treating samples with stereoisomers of simple nutrients (ie. small amino acids or sugars) prior to DS, may also help in distinguishing biological from sterile samples. Finally, we have developed nonlinear dielectric response techniques, which will also be tested for their ability to differentiate living from sterile material.

Atmospheric Electron-Induced X-ray Spectrometer (AEXS) Development

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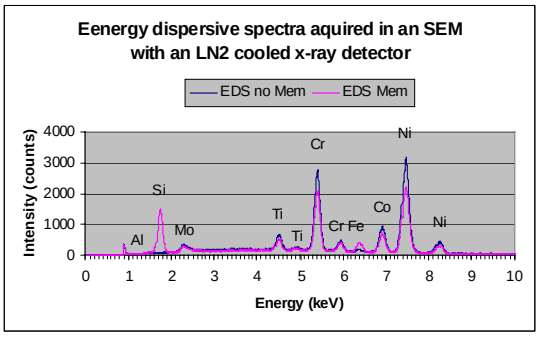
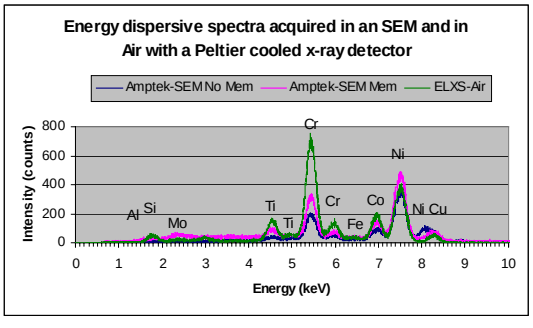
The AEXS is a miniature, low mass, low energy consumption, portable instrument for elemental surface analysis *in situ,* with high spatial resolution (sub mm-size) and short (~minute) spectrum acquisition times. The AEXS excites characteristic x-ray fluorescence (XRF) using an electron beam from samples in ambient atmosphere, requiring no sample handling; the excitation is made possible by isolating the vacuum of the electron column from the electron excitation is very effective in generating XRF, and a high intensity beam can be focused into a small spot. The rapidity of the spectrum acquisition (within minutes) and consequently low energy consumption (~50 Joules per acquired spectrum) will enable mapping the elemental composition of soil or rock surfaces in either a spot analysis mode (several 1 mm^2 irradiated spots), or in a survey mode (1-2cm^2 scanned area).

The AEXS, located on a rover or lander arm addresses the science needs of Mars Scout and MSL missions by providing a rapid means for elemental characterization of soil or rock surfaces. An instrument suite that includes a catholuminescence (CL) detector would permit to also detect CL spectra, thus being able to detect water-deposited minerals such as carbonates, which commonly exhibit CL. The AEXS also addresses the needs of a future MSR mission via its ability to rapidly screen potential samples for caching and return. The high flux of electrons could be used for ionizing gaseous species for further *in situ* analysis by other instruments such as mass spectrometers.





We report on our progress to date. Using an encapsulated 10 keV electron source, we have acquired XRF spectra from samples in ambient atmosphere, and compared them successfully with spectra obtained in an SEM using the EDS and Amptek detectors with and without the intervening membrane from several samples. Using a CL detector, we have obtained different CL spectra from different areas on silicate minerals, some of which were altered by microbial activities. The spectra were compared with EDS spectra analysis. To improve the excitation efficiency and reliability, we have adapted and vacuum-isolated a 20keV electron gun. The 20keV source is our baseline design for adaptation onto a rover arm, and is being used to acquire data in a vacuum chamber at Mars atmosphere pressure.

Field Tests of the Mars Oxidant Instrument (MOI) A. P. Zent, NASA Ames Research Center, R. C. Quinn, SETI Institute, F. G. Grunthaner, Jet Propulsion Lab, P. Ehrenfreund, Leiden Observatory

An implementation of the Mars Oxidant Instrument (MOI) was field tested at the Yungay Field Station (24°S), in arid core of the Atacama Desert, Chile, during February, 2004 Gonzalez-Navarro et al. (2003) demonstrated that the quantity and diversity of bacteria increase as a function of local water availability in the Atacama and that for some soil samples from the Yungay region, no bacteria could be isolated. Flash pyrolysis GCMS analysis of soils collected from these regions revealed extremely low levels of organic matter. The existence of organic-depleted, near-sterile soil offers an interesting Earth analog of the Martian surface material.

The Mars Oxidant Instrument, in the configuration tested here, records the DC resistivity of thin test-films exposed to the environment. MOI deployed identical film sets in configurations that permitted exposure to a) UV, b) dust, c) both (unfiltered) and d) neither. It was deployed to assess the chemical processes occurring in the environment.

Distinct reaction patterns are observed for each of three different Au-film sensor deployment modes. Reactivity is highest and reaction kinetics are fastest for sensors exposed to atmospheric dust. For the UV+Dust sensors, reaction proceeds after a twenty four hour dust accumulation period and an increase in atmospheric humidity at night. The observed response is consistent with dry-acid (H_2SO_4 and HCl) deposition on the sensors. Atmospheric sensors shielded from dust did not exhibit this response. Sensors exposed to soil displayed slower reaction kinetics due to buffering of humidity by the soil.

Non-aqueous pH measurements of the surface crusts in the confirmed very low pH in the presence of trace H_2O (Quinn et al., In prep). Atmospheric R_H reached > 90% overnight, occasionally approaching unity. Thin H_2O films solvate H_2SO_4 and HCl, attacking both test organic compounds and the underlying Au conducting films.

The majority of dust in the vicinity is gypsum, and the composition of the acidic soil components is likely dominated by H_2SO_4. The use of the MOI sensors in different configurations modes, as demonstrated here, discriminates between oxidant reaction pathways and will allow rapid characterization of environmental chemical processes on Mars.

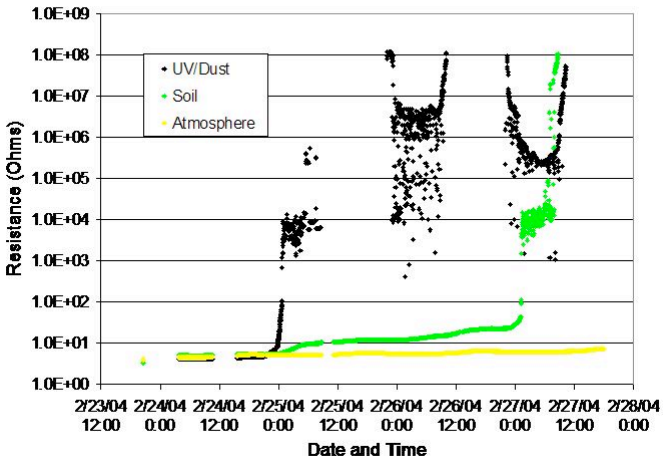


Figure 1. Resistance changes in thin-gold films as a function of time and deployment mode, Chemical modification of the films was induced by a combination of dust and water exposure. Films exposed only to the atmosphere remained essentially unchanged. In sensors exposed to soil or dust, chemical modification was triggered by solvation of acidic soil components due to changes in humidity.